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THE EFFICIENCY OF GAUZE MASKS IN THE PROTECTION OF RABBITS AGAINST THE INHALATION OF DROPLET NUCLEI OF TUBERCLE BACILLI[1, 2, 3]

MAX B. LURIE AND SAMUEL ABRAMSON[4]

INTRODUCTION

Although there is no certain knowledge as to the size of the infectious particles concerned in the origin of human inhalation tuberculosis, there is considerable circumstantial evidence to suggest that the entities involved are much smaller than the limits of ordinary visibility (1). As is well known, primary pulmonary tuberculosis in man takes root not in the upper respiratory passages but deep in the lung parenchyma, usually beneath the pleura. It is reasonable to assume, therefore, that the effective pathogenic units are smaller than the lumina of the terminal bronchioles. That the tubercle-bacilli-containing-particles responsible for the inception of naturally acquired air-borne pulmonary tuberculosis in rabbits are of microscopic dimensions was indicated by the effect of ultraviolet irradiation of the air of a room contaminated by tubercle bacilli derived from tuberculous animals. Such irradiation protected rabbits from an airborne contagion that caused progressive tuberculosis in 73 per cent of animals of the same genetic resistance to tuberculosis exposed to the same contagion for the same period of one year in an unirradiated room (2). As the ventilation of the irradiated room was such that some droplet nuclei of tubercle bacilli floating in the air were exposed for as short a period as only one second to 2537 Å ultraviolet energy of an average maximum intensity of 200 μ W per cm^2 before their inhalation by the exposed rabbits, and as these particles were sterilized, as shown by failure of the animals to develop tuberculosis, it is likely that the infectious units are microscopic.

Therefore, it was not at all certain whether gauze masks with pores of relatively large magnitude, such as may be worn by individuals exposed to airborne contagion of human tuberculosis, would filter out the highly dangerous invisible droplet nuclei of tubercle bacilli. There is some evidence that six layer gauze masks, especially after repeated washing, will remove bacteria floating in the air and yet will not interfere significantly with the free passage of air necessary for respiration (3).

Miss Esta H. McNett, of the Veterans Administration, who devoted many years of effort toward devising means of protecting nurses engaged in the care of tuberculous patients, has designed a six-layered gauze mask to be worn by

[1] From the Henry Phipps Institute of the University of Pennsylvania, Philadelphia, Pennsylvania.

[2] Aided by grants from the Commonwealth Fund, the Ella Sachs Plotz Foundation and the National Tuberculosis Association.

[3] Presented before the Conference on Tuberculous Nursing, as part of the symposium on *Ways to Enhance the Value of Services*, at the 44th annual meeting of the National Tuberculosis Association, New York, New York, June 14, 1948.

[4] Tuberculosis Control Division, Public Health Service, Federal Security Agency.

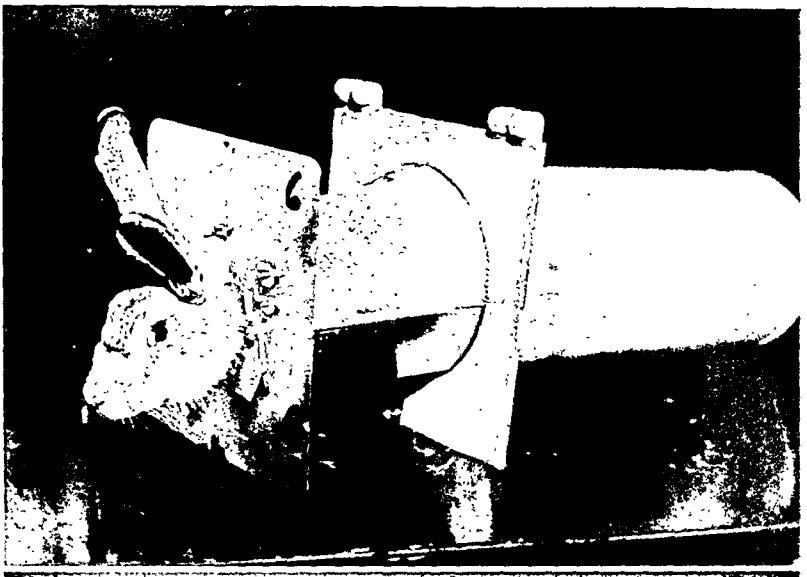

Fig. 1

Fig. 2

Fig. 3

Plate I

6 rabbits were exposed simultaneously, 4 without masks and 2 with masks. Two of the 4 rabbits without masks were killed immediately after exposure in order to determine the number of units of living tubercle bacilli retained by the lungs. The lungs were removed under sterile conditions, the bronchi cut at their junction with the lung parenchyma and weighed. The specimens were ground in sterilized, chilled Waring blendors with 9 volumes of cold M/15 Na_2HPo_4 for four minutes. Aliquot portions of this lung tissue brei, as well as portions of the same suspensions treated with 6 per cent sulfuric acid, were planted on a modified Löwenstein's medium (5). The number of colonies derived from a known weight of lung parenchyma indicated the total number of viable bacilli present in both lungs. This procedure makes possible the determination of the minimum number of bacillary units retained by the lung parenchyma.

The remaining 4 rabbits were allowed to live until the control unmasked rabbits developed strong tuberculin reactions. All of the 4 rabbits were then killed and the number of primary tubercles present in their lungs was accurately determined by excising each individual primary focus, except as otherwise indicated.

The results are presented in table 1. In columns 3 and 4 of the table are listed the number of bacillary units isolated from the lungs of rabbits without masks which were killed immediately after exposure. With few exceptions, considerable numbers of tubercle bacilli were isolated in each instance. These ranged from 30 to over 1,000 and averaged 275 tubercle bacilli units per rabbit. Every one of the rabbits that survived four or more weeks, exposed without masks, developed primary tubercles which ranged in number from 3 to 136 and averaged 28 tubercles per animal. Twelve of the 20 masked rabbits which were simultaneously exposed, and killed at the same time as the controls, showed no grossly visible tubercles. Two such examples of the effectiveness of the gauze mask in the protection against tuberculosis are illustrated in figures 4 and 5. The remaining masked rabbits were not completely protected. While the rabbits without masks exposed simultaneously with the masked animals developed from 3 to 136 tubercles and averaged about 31 per rabbit, the masked rabbits developed from 1 to 9 tubercles and averaged less than 4 per rabbit. Thus, even in the instances where the masks failed to afford complete protection, they prevented the inhalation of infectious particles to such an extent that the number of tubercles developed was reduced eightfold.

If the total number of tubercles developed by the 18 rabbits without masks is compared to the total number of tubercles developed by the 19 masked rabbits simultaneously exposed, the resulting values are 28 tubercles per rabbit for unmasked animals and 1.4 tubercles per rabbit for the masked animals. This represents a 95 per cent efficiency for the mask in suppressing grossly visible primary pulmonary tubercles.

In an effort to eliminate the role of variations in individual resistance to tuberculosis, another experiment was done. Six inbred rabbits, of the cross between the A and D families (6), were exposed simultaneously to the inhalation of large

numbers of droplet nuclei of tubercle bacilli of the Ravenel strain. Three were masked and three were without masks. The results are presented in table 2.

TABLE 1

Efficiency of gauze masks in the protection of rabbits against the inhalation of droplet nuclei of tubercle bacilli

EXPERIMENT NUMBER	RAVENEL NUMBER AND TIME SAID EXPOSED TO EXPERIMENT	CONTROL				MASKED	
		Number of tubercle bacilli cultured from both lungs		Number of tubercles in both lungs		Number of tubercles in both lungs	
		Rabbit 1	Rabbit 2	Rabbit 3	Rabbit 4	Rabbit 5	Rabbit 6
1	No. 1, 0 No. 2, 0	—	29	4	3	1	0*
2	No. 1, 1 No. 2, 1	—	75	3	6	1	0
3	No. 1, 2 No. 2, 2	255	317	59‡	41	0?‡	0
4	No. 2, 3 No. 3, 0	—	—	**	3	0	0
5	No. 2, 4 No. 3, 1	33	29	12‡	4	0?‡	0
6	No. 5, 0 No. 6, 0	720	—	21	23	***	0
7	No. 7, 0 No. 8, 0	116	—	7	****	3	0
8	No. 9, 0 No. 10, 0	54	1,027	27	32	0	3
9	No. 1, 3 No. 2, 5	131	530	60	15	0	1
10	No. 1, 4 No. 2, 6	222	287	54	136	9	9

* Died one day after exposure; no tubercle bacilli could be demonstrated in the lungs by culture or guinea pig inoculation.

** Died one day after exposure; 47 tubercle bacilli cultured from lungs.

*** Died immediately after exposure; tubercle bacilli demonstrated in lungs by guinea pig inoculation.

**** Died four days after exposure; 172 tubercle bacilli cultured from lungs.

‡ Number of tubercles determined by careful inspection and palpation.

The number of bacilli inhaled was such that the unprotected rabbits developed 126, 219, and 223 tubercles, respectively, in the lungs five weeks after exposure. On the other hand, of the masked rabbits, two, which were killed simultaneously

FIG. 4

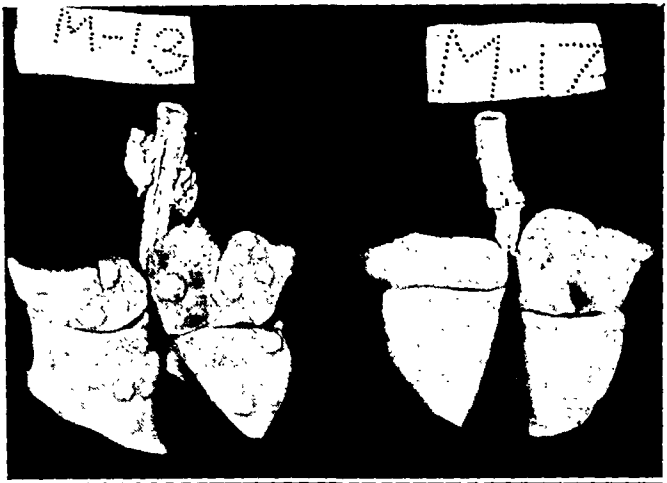

FIG. 5

FIG. 4. The lungs of rabbit M-13 (unmasked) and of rabbit M-17 (masked), at autopsy thirty-four days after a simultaneous exposure to droplet nuclei of virulent bovine type tubercle bacilli. Fifty-nine primary tubercles were counted in the lungs of the control rabbit (M-13). No tubercles were seen in the masked animal (M-17).

FIG. 5. The lungs of rabbit M-25 (unmasked), and of M-26 (masked) at autopsy forty-four days after a simultaneous exposure to droplet nuclei of virulent bovine type tubercle bacilli. Twelve primary tubercles were counted in the lungs of the control rabbit M-25. No tubercles were seen in the masked animal (M-26).

TABLE 2

Efficiency of gauze masks in the protection of rabbits against the inhalation of droplet nuclei of tubercle bacilli—Experiment 11

	CONTROL			MASKED		
RABBIT NUMBER	A D 2-31	A D 3-9	A D 3-5	A D 3-4	A D 2-37	A D 2-28
Mask number and times used previous to experiment	none	none	none	No. 3, 3	No. 2, 7	No. 1, 5
Number of tubercles in both lungs	223	126	219	60	7	0*

* Rabbit died 3 days after exposure; no tubercle bacilli could be obtained from the lungs by culture or guinea pig inoculation.

with the control rabbits, had 60 and 7 tubercles, respectively, in their lungs. One rabbit died three days after exposure. No tubercle bacilli could be demonstrated either by culture or guinea pig inoculation in this rabbit's lung. Thus, the efficiency of the masks in this experiment, based on the standards outlined above, was approximately 88 per cent, which is not materially different from that in the previous 10 experiments.

Addition of the results obtained with these 6 rabbits exposed to massive numbers of tubercle bacilli to those listed in table 1 reveals an average of 51 tubercles per rabbit in the unmasked animals and 4.3 tubercles per rabbit in the masked animals, or a mask efficiency of approximately 92 per cent.

In column 2 of table 1 are given the serial numbers of the masks used for each rabbit. In each experiment the number of the mask listed first was used for rabbit 5 and the mask listed second was used for rabbit 6. In the same column are indicated the number of times each mask was used and the number of times it had been washed and autoclaved before further use. In some experiments the masks consisted of six layers of gauze, in others they were three-layered. It will be noted that no consistent difference in the efficiency of these masks was found under these varying conditions.

DISCUSSION

It has been found in this study that, if all the air respired by rabbits exposed to the inhalation of droplet nuclei of virulent bovine tubercle bacilli passes through three or six layer gauze masks, there is a 90 to 95 per cent reduction in the incidence of primary pulmonary tuberculous foci which develop within five weeks. It would follow, therefore, that, if the respired air contains but a few bacilli, the masked animal will usually be protected from an otherwise fatal infection. Indeed, 12 of 20 masked animals were completely protected against air-borne contagion of such intensity that from 29 to 1,027 tubercle bacilli units were deposited in the lungs of simultaneously exposed unmasked rabbits.

Measurements of the thread diameters and interthread spaces of these masks showed (7) that the superimposition of three to six layers of this material would occlude practically all of the spaces and in this way explain the results of the experiments.

It is not unreasonable to assume that the depth and force of the inspiration of the rabbits during their exposure, which varied with each animal, may be a determining factor. The deeper and more forceful their inspiration, the less effective may be the filtering capacity of the masks. This remains to be determined. There are many other facts which can be ascertained relative to this problem by the methods described in this paper.

In applying these data to the protection of human beings one must be extremely guarded. The masks were so applied to the rabbits that all of the respired air passed through the masks. To be equally effective for human beings exposed to air-borne infection of tuberculosis, the mask must be worn in an equally effective manner. It would seem that this offers no insurmountable difficulty. If the frame into which the mask fitted could be constructed of pliable material

which could be accurately applied to the contour of the individual's face around the nose and mouth, and if this contact could be made airtight, there is no reason to believe that the mask could not effectively filter out the dangerous invisible particles that are concerned with the inception of pulmonary tuberculosis. The masks protected rabbits from air populated with droplet nuclei of tubercle bacilli to a degree that would be rarely, if ever, found in the air respired by human beings. For, as is well known, human primary tuberculosis usually originates as a single pulmonary focus, whereas the unmasked rabbits in these experiments developed an average of 51 primary tubercles. Nevertheless, it seems reasonable to advise persons wearing masks for this purpose to refrain from deep inspiration as much as possible as it is not unlikely that forceful suction produced by deep inhalation may diminish the filtering efficiency of the masks.

Conversely, as has been shown by Jennison (8), masks worn by coughing patients can hardly be expected to retain the invisible droplet nuclei containing tubercle bacilli propelled through them by the often extremely forceful expiratory chest movements during fits of coughing.

SUMMARY

Under the conditions of these experiments, from 90 to 95 per cent of pathogenic droplet nuclei of virulent bovine tubercle in the respired air can be removed by gauze masks properly worn by rabbits during quiet breathing of heavily infected air.

A method is described by which much pertinent information in regard to this problem can be acquired.

The application of these data to the wearing of masks by human beings exposed to air-borne tuberculous contagion is discussed.

SUMARIO

La Eficacia de las Máscaras de Gasa para la Protección de los Conejos contra la Inhalación de Núcleos de Bacilos Tuberculosos en Gotillas

En las condiciones de los experimentos descritos, pueden eliminarse del aire respirado de 90 a 95 por ciento de los núcleos en gotillas patógenas de bacilos bovinos virulentos, por medio de máscaras de gasa debidamente puestas en conejos, durante la respiración tranquila de aire intensamente infectado.

Descríbese un método que permite adquirir mucha información pertinente acerca de este problema.

Se discute la aplicación de los datos obtenidos al uso de máscaras por los seres humanos expuestos al contagio tuberculoso por vía aérea.

Acknowledgment

We are greatly indebted to Miss Fannie Eshleman for assistance in devising a mask suitable for rabbits and to Mr. Peter Zappasodi for the photographs for this paper.

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PULMONARY RESECTION IN THE TREATMENT OF TUBERCULOSIS[1, 2]

JOSEPH W. GALE, HELEN A. DICKIE, AND ANTHONY R. CURRERI

INTRODUCTION

Surgical collapse therapy in the treatment of pulmonary tuberculosis has progressed to the stage where indications for operative interference have been well established and final results can be predicted with more than a reasonable degree of assurance. There remains, however, a certain group of patients who have failed to convert after the use of conventional methods of surgical collapse and who need further surgical measures to rid them of active tuberculous foci. Therefore, it is not surprising that pulmonary resection, so successful in its application in the treatment of various nontuberculous lesions, should have been tried in these cases.

In 1942 Thornton and Adams (1) collected and reported the cases of resection for pulmonary tuberculosis. There were 29 cases of pneumonectomy with a mortality of 45 per cent and 46 cases of lobectomy with a mortality of 25 per cent. The group varied so greatly that it was impossible to evaluate the results.

Recently Janes (3), Maier and Klopstock (5), Overholt *et al.* (7), Sweet (21), Bailey (9), and Clagett and Seybold (14) have reported larger individually controlled groups. The results of these studies show a more encouraging picture as to early operative mortality and end results. The improvement in the results is to be expected in view of the refinements in surgical technique employed in dealing with the hilar structures. More recently, however, Alexander (10) has compiled the largest group of cases (411) and found that only 45 per cent are apparently well and without tubercle bacilli in their sputum, 25.5 per cent are dead, and 29.5 per cent of the whole group have active uncontrolled disease. A study of such a large group demonstrates quite clearly that resection, as used to date, is not the answer to the problem.

The present report deals with 80 resections performed at the State of Wisconsin General Hospital over a period of three years. The patients were referred from various sanatoriums throughout Wisconsin. Most of them had had some type of collapse therapy prior to admission and they represented a group in need of further treatment. After thorough study they were brought before the combined medical and surgical staffs concerned with the treatment of pulmonary tuberculosis. Further measures were discussed and decided upon. In this group of patients such a concerted study was preferred to the decisions of only one or two individuals.

[1] From the Departments of Surgery and Medicine, University of Wisconsin Medical School, Madison, Wisconsin.

[2] Presented before the Medical Section, as part of the *Symposium on Surgery—Trends—Late Results*, at the 44th Annual Meeting of the National Tuberculosis Association, New York, New York, June 16, 1948.

OPERATION

The operative technique employed was quite similar to that used by others. The patient was placed on his less involved side. If there was excessive bronchial secretion, a bronchoscopy was done and the offending side plugged with a Tampax pack. The anesthetic agent varied and consisted for the majority of the patients of cyclopropane induction followed by ether. The incision most recently employed was that used for first-stage thoracoplasty. This is of definite advantage if thoracoplasty is to be performed at the same sitting. The sixth or seventh rib was resected after the intercostal nerves of the resected and adjoining ribs were crushed to reduce postoperative pain. Following resection, one gram of streptomycin and 100,000 units of penicillin were introduced into the pleural space. No drainage was used following pneumonectomy but in all cases of lobectomy drainage was instituted and maintained for at least forty-eight hours. If additional air or fluid accumulated after removal of the tube, aspiration was done promptly to promote complete expansion of the remaining lobe or lobes. The intrapleural pressure was kept negative following pneumonectomy. Thoracentesis was repeated every other day. Fluid was withdrawn, one-half gram of streptomycin instilled, and the pleural pressure readjusted. These procedures were continued until the pleural space was obliterated and fluid no longer accumulated. Patients who had had a previous thoracoplasty were observed closely because the small pleural space following pneumonectomy reacted quickly, and at times violently, to minor pressure changes. It is felt that the early obliteration of the pleural space has been most valuable in preventing the development of early and late tuberculous empyema. Upper stage thoracoplasty was done at the time of resection if the condition of the patient permitted. If not advisable then, it was carried out as soon afterwards as possible.

Phrenicectomy was done routinely following pneumonectomy but the function of the diaphragm was preserved following lobectomy to ensure normal activity of the remaining lobe and as a guard against atelectasis. Bronchoscopy was used immediately postoperatively only if secretions had been troublesome during the operation. Blood transfusions were used freely during the operation and throughout the postoperative period. The patient was placed in the Trendelenberg position for the first twenty-four hours. Frequent vigorous changes in position were mandatory. Subcutaneous and intravenous fluids were given until the patient was able to maintain his fluid balance. The patient was returned to the sanatorium as soon as he was judged fit to travel by ambulance.

INDICATIONS

Many factors enter into the consideration of any patient as a candidate for resection. Nevertheless, certain broad indications must be adopted in order to have a starting point. The indications used in this group of patients conform surprisingly well to those followed by a number of other surgeons (table 1).

Thoracoplasty failure: The group of patients in whom thorocoplasty had failed represented the chief indication for resection (tables 2 and 3). There were 41 such cases (51.25 per cent). Twenty-six (32.5 per cent) had pneumonectomy and fifteen (18.75 per cent) had lobectomy. Frequently there were other important lesions present, some of which may be considered as definite indications for resection; but these were considered secondary if a previous thoracoplasty had been performed. Seventeen patients had received pneumothorax, 12 had stenosis of the lobar or stem bronchus, 7 showed severe bronchiectasis, 6 had repeated

pulmonary hemorrhages, and 2 had empyema, one tuberculous, the other a mixed infection. The lobectomy group contained 9 with bronchiectasis, 8 with cavity or cavities, 3 with atelectasis, 2 with recurrent hemoptysis, and one with lobar stenosis.

TABLE 1

Broad indications for resection (various surgeons)

NAME	THORACOPLASTY FAILURE	BRONCHOSTENOSIS	TUBERCULOMA	LOWER LOBE AND BASAL CAVITIES	TUBERCULOUS BRONCHIECTASIS	GIANT CAVITY DESTROYED LUNG	PNEUMOTHORAX FAILURE
Adams (2)	+		+	+	+	+	
Alexander (11)	Patients for whom collapse therapy is unsuitable or for whom it has been tried unsuccessfully						
Clagett (13)	+	+	+		+	+	
Dolley (12)	+	+		+	+	Tension cavity	
Janes (4)	+	+	+	If pneumothorax or phrenic surgery fails			+
Jones (15)	+	+	+	+	+		+
Kinsella (16)	+	+			+		
Maier (6)	+	+	+	+	+	+	+
O'Brien (17)	+	Over 70 per cent	+	+	Does not respond to streptomycin or is very extensive.		+
Ochsner (18)	+	+	+				
Overholt (8)	+	+	Of size	+	+	+	
Samson (19)	+	+	+	+	+	+	
Strieder (20)	+	+	+		+	Tension cavity	
Sweet (22)	+	+	+	+	Distal to obstruction.		
Woodruff (23)	+	High grade	+	+			
Authors	+	+	+	+	+	+	+

Bronchostenosis: The second most common indication for resection was bronchostenosis, which was present in 16 cases (20 per cent). Thirteen (16.25 per cent) of this group had pneumonectomy and 3 (3.75 per cent) lobectomy (tables 4 and 5). In the pneumonectomy group there were 5 cases of atelectasis,

TABLE 2

Thoracoplasty failure (lobectomy)

PATIENT, AGE, SEX	STAGE DURATION PREVIOUS THERAPY	INVOLVEMENT	LOBECTOMY DATE	POSTOPERATIVE COURSE	PRESENT STATUS
F. N. 52 M	II, 5 years Thoracoplasty	Bronchiectasis RU and ML, pneumonitis	RU and ML 4-24-45	Continued improvement	9-1-47 "negative" sputum, full time work
C. P. 33 F	III, 3 years Phrenic surgery, thoracoplasty, revision	Cavity RUL, bronchiectasis, atelectasis, bronchitis	RUL 5-23-45	Uneventful	January 1948 "negative" sputum, working as nurse, symptom free
E. P. 27 F	III, 3 years Pneumothorax, pneumoperitoneum, thoracoplasty	Cavity LUL	LUL 5-24-45	Died in October 1947. Spread to contralateral lung	Dead
K. R. 24 M	III, 5 years Pneumothorax, thoracoplasty	Bronchiectasis LUL with hemoptysis	LUL 10-1-46	Severe dyspnea for 4 weeks because of low respiratory reserve	9-5-47 "negative" sputum, at home, working, roentgenograms stable
A. B. 39 F	III, 8 years Phrenic surgery, thoracoplasty, revision	Bronchiectasis and cavity RUL, stenosis UL bronchus	RU and ML 11-30-46	Atelectasis RLL for 4 days	9-1-47 "negative" sputum, at home, continued well
L. B. 23 M	III, 6 years Thoracoplasty	Atelectasis RUL with bronchiectasis	RU and ML 2-24-47	Vertigo due to streptomycin	9-24-47 "negative" sputum, 12-20-47 "negative" sputum, rehabilitation at sanatorium
A. W. 45 M	III, 4 years Pneumothorax, thoracoplasty	RU and ML Bronchiectasis	RU and ML 2-26-47	Uneventful	9-1-47 "negative" sputum, no change January 1948, 4-15-48 "negative" sputum
V. O. 26 M	II, 5 years Phrenic surgery, thoracoplasty	Bronchiectasis RUL	RUL 3-27-47	Developed cavity in contralateral upper lobe November 1947 closed and remained closed	9-1-47 "negative" sputum, 1-19-48 "negative" sputum
A. W. 33 M	III, 9 years Thoracoplasty, revision, pneumothorax on opposite side	Cavity RUL	RU and ML 7-18-47	Dyspnea and palpitation, responded slowly	8-14-47 sputum positive for tubercle bacilli, 4-15-48 sputum positive for tubercle bacilli
R. B. 44 M	III, 3 years Thoracoplasty for tension cavity	Large cavity RUL, hemoptysis, red bronchus	Ru and ML 9-17-47	Slow continued improvement	10-29-47 sputum positive for tubercle bacilli, 3-1-48 "negative" sputum following three cultures

TABLE 2—*Concluded*

PATIENT, AGE, SEX	STAGE DURATION PREVIOUS THERAPY	INVOLVEMENT	LOBECTOMY DATE	POSTOPERATIVE COURSE	PRESENT STATUS
H. D. 27 F	III, 5 years Thoracoplasty	Multiple cavities RUL, bronchiectasis	RUL 10-15-47	Atelectasis of M and L immediately postoperatively subsided	3-9-48 "negative" sputum, roentgenograms stable
M. A. 38 F	III, 12 years Thoracoplasty	Cavity RUL	RUL 10-24-47	Reactivation of tuberculosis otitis media	11-8-47 "negative" sputum, 4-1-48 "positive" sputum, otherwise excellent
H. S. 52 M	III, 3 years Thoracoplasty	Cavity and bronchiectasis RUL	RUL 10-24-47	Chest has remained stable	11-18-47 "negative" sputum, 4-15-48 "negative" sputum
I. M. O. 36 F	III, 9 years Pneumothorax, thoracoplasty	Atelectasis and fibrosis LUL	LUL 12-22-47	Auditory and visual hallucinations	4-15-48 "negative" sputum
E. L. 43 F	III, 15 years Thoracoplasty	Large thin-walled at medial side of left apex	LUL 2-10-48	Uneventful	4-15-48 sputum "negative" on smear

2 with repeated hemoptysis, and 3 with cavities. One patient (M.M.) did not have a thoracoplasty following lobectomy and developed a "spread" in the apex of the right lower lobe eight months later. A thoracoplasty was performed to control the "spread."

Tuberculous bronchiectasis: The third most common indication, tuberculous bronchiectasis, concerned only 8 cases (10 per cent). All patients in this group had lobectomy with the removal of one or two lobes. Two had cavitation of the involved lobe, in addition one had repeated hemoptyses, and one a contralateral pneumothorax at the time of resection. Three patients had thoracoplasty performed at the time of lobectomy, and three soon thereafter. One patient (M.M.) had a segmental resection of the apex of the lower lobe. This was the only time segmental resection was done in the entire series. One postoperative death occurred.

Lower lobe lesions: The group of lower lobe lesions consisted of 4 cases (table 7). The patient (J.DeS.) might have been placed in another group but the existence of a tuberculoma in the lower lobe would appear to justify his being included here.

Pneumothorax failure: Four cases with unsuccessful pneumothoraces have been included (table 8). One patient (C.S.) suffered a spontaneous collapse as the first recognized symptom early in the course of his disease; this resulted in a tuberculous empyema and a nonexpansile lung. Two patients had nonexpansile lungs. One patient had a thick-walled cavity in a well collapsed right upper lobe. Two had lobectomy, one with decortication and expansion of the remaining lobe, while 2 had pneumonectomy. All received a later thoracoplasty.

Destroyed lung: Three patients had far advanced lesions with widespread

TABLE 3

Thoracoplasty failure (pneumonectomy)

PATIENT, AGE, SEX	STAGE DURATION PREVIOUS THERAPY	INVOLVEMENT	PNEUMONECTOMY DATE	POSTOPERATIVE COURSE	PRESENT STATUS
H. S. 36 F	III, 8 years Sanatorium, phrenic surgery, pneumothorax, thoracoplasty	Stenosis LUL bronchus, atelectasis of UL	Left 12-7-44	Coughed up silk sutures 6 months later; spread to right lung which cleared; reactivation right lung February 1948	11-25-47 sputum "positive," 4-15-48 sputum "positive"
I. M. 36 F	III, 5 years Sanatorium, phrenic surgery, thoracoplasty	Extensive cavitation, (Destroyed lung)	Right 5-21-45	Uneventful	3-1-47 sputum "negative," continued well, 4-15-48 full time work
L. W. 35 M	III, 5 years Pneumothorax, thoracoplasty, revision	Persistent cavity, hemoptysis	Right 6-29-45	Hemothorax, postoperatively controlled	9-1-47 sputum "negatives," TB sinus chest wall, Reoperated 4-15-48 negative)
S. L. 38 F	III, 15 years Phrenic surgery, thoracoplasty, revision	Bronchiectasis, high grade stenosis right stem bronchus	Right 10-26-45	Uneventful	4-15-48 sputum "negative," part time work, low pulmonary reserve as before operation
T. E. 46 M	III, 4 years Pneumothorax, thoracoplasty	Stenosis RUL bronchus, mixed infection empyema	Right 4-30-46	Immediate reactivation on left with effusion, cleared with streptomycin	3-9-48 sputum "negative"
A. K. 40 F	II, 10 years Phrenic surgery, pneumothorax, thoracoplasty	RUL cavitation, bronchiectasis, red bronchus	Right 6-14-46	Uneventful	9-1-47 sputum "negative," at home; January 1948 continues well; excellent
E. L. 25 F	III, 7 years Pneumothorax, phrenic surgery, thoracoplasty	Atelectasis with tuberculous bronchitis	Right 6-20-46	Uneventful	9-1-47 sputum "negative," full time work, continues well 4-15-48
V. K. 20 F	III, 6 years Pneumothorax, thoracoplasty	RUL bronchial stenosis, atelectasis of right lung with cavity	Right 7-2-46	Ulceration of bronchial stump, visible silk sutures, to be removed	12-1-47 sputum "positive," should clear with removal of silk sutures
L. K. 29 M	III, 4 years Pneumothorax, thoracoplasty	Stenosis of left stem bronchus; tuberculous bronchitis, active	Left 7-3-46	Reactivation on right August 1947 after discharge from sanatorium, cleared with streptomycin	10-2-47 sputum "positive," 4-15-48 sputum "negative"
A. B. 38 M	III, 13 years Phrenic surgery, pneumothorax, thoracoplasty	Cavitation LUL, stenosis stem bronchus, suppurative pneumonitis	Left 2-10-47	Continued improvement	9-1-47 sputum "positive," 1-24-48 sputum "negative," 2-28-48 sputum "negative," at home
E. F. 54 M	III, 9 years Thoracoplasty	Persistent cavitation, intermittent hemoptysis	Right 3-11-47	Died suddenly 9-7-47 of coronary occlusion	Sputum "negative" 9-1-47 six days before death

TABLE 3—*Continued*

PATIENT, AGE, SEX	STAGE DURATION PREVIOUS THERAPY	INVOLVEMENT	PNEUMONECTOMY DATE	POSTOPERATIVE COURSE	PRESENT STATUS
F. S. 27 F	III, 2 years Thoracoplasty	Fibrocaseous tuberculosis with cavitation, bronchiectasis and increasing atelectasis	Right 3-11-47	Reactivation at left apex, streptomycin for 90 days halted disease, now stable	9-1-47 sputum "positive," 2-28-48 sputum "negative"
C. R. 22 F	III, 6 years Pneumothorax, thoracoplasty	Fibrocaseous with cavitation, stenosis right stem bronchus	Right 3-25-47	Ulceration of bronchial stump 10-1-47, cleared following streptomycin for 150 days	9-1-47 sputum "positive," 2-15-48 sputum "negative"
S. M. P. 44 F	III, 19 years Thoracoplasty	Pyogenic infection superimposed on cavernous lesion, uncontrolled cough	Left 5-2-47	Streptomycin reaction, cyanosis, dyspnea; a respiratory cripple	9-1-47 sputum "positive," dyspnea improving, died 2-28-48 cardiac failure
M. K. 41 F	III, 6 years Phrenic surgery, pneumothorax, pneumoperitoneum, thoracoplasty	Bronchiectasis RU and ML, repeated hemoptysis	Right 5-10-47	Complicating peptic ulcer, bleeding; lung stable	10-1-47 sputum "negative;" continues well 4-15-48
N. D. P. 44 M	III, 7 years Pneumothorax, pneumoperitoneum, phrenic surgery, thoracoplasty	Extensive lesions throughout lung, active bronchitis cleared with streptomycin	Left 7-28-47	Convalescence satisfactory, now in rehabilitation in sanatorium	1-5-48 sputum "negative"
A. J. 34 F	III, 8 years Thoracoplasty, revision	Stenosis RUL bronchus, cavitation U and LL, bronchiectasis	Right 9-11-47	Left lung has remained stable	4-15-48 sputum "negative"
I. W. 44 F	III, 5 years Permanent, phrenic surgery, thoracoplasty	Pinpoint stenosis, suppurative pneumonitis, hemoptysis, bronchiectasis	Left 9-15-47	Right lung continued to remain quiescent	1-4-48 sputum "negative," on rehabilitation in sanatorium
E. T. 28 F	II, 7 years Phrenic surgery, thoracoplasty	Fibrocaseous tuberculosis, bronchiectasis, repeated hemoptysis	Left 10-10-47	Rapid convalescence	12-20-47 sputum "negative," right lung normal 3-24-48
F. R. 21 M	III, 2 years Phrenic surgery, pneumoperitoneum, thoracoplasty	Multiple bronchiectatic cavitation, repeated hemoptysis	Left 10-16-47	Has remained quiescent since operation	Six sputum "negative" cultures since operation
M. G. 28 F	III, 8 years Phrenic surgery, thoracoplasty	Stenosis left stem bronchus, many bronchiectatic cavities, cavities in LL	Left 11-28-47	Streptomycin started 3-4-48, no evidence of spread or reactivation	3-24-48 sputum "positive"
L. Y. 21 F	III, 4 years Thoracoplasty	Multiple cavities, bronchiectasis	Left 12-3-47	Right lung has continued to remain stable	1-2-48 sputum "negative"

TABLE 3—*Concluded*

PATIENT, AGE, SEX	STAGE DURATION PREVIOUS THERAPY	INVOLVEMENT	PNEUMONECTOMY DATE	POSTOPERATIVE COURSE	PRESENT STATUS
C. L. 48 M	III, 9 years Pneumothorax, oleothorax, thoracoplasty, streptomycin	Pinpoint stenosis stem bronchus, cavity at apex, tuberculous empyema	Left 12-8-47	Condition has gradually improved	2-8-48 sputum "negative"
M. M. 22 F	III, 3 years Pneumothorax, promine, thoracoplasty	Bronchiectasis, stricture LL bronchus, atelectasis	Left 12-18-47	Patent ductus ligated at operation	1-4-48 sputum "negative," right lung stable
M. H. 26 F	III, 5 years Thoracoplasty, 1945	Swelling of right stem bronchus, bronchial "spread"	Right 2-4-48	Continued improvement	4-15-48 sputum "negative"
D. W. 30 F	III, 5 years Phrenic surgery, right 1944; thoracoplasty, right 1946	Tuberculous granulations in right stem bronchus, mucopurulent secretions	Right 2-12-48	Continued improvement	4-15-48 sputum "negative"

TABLE 4

Bronchial stenosis (lobectomy)

PATIENT, AGE, SEX	STAGE DURATION PREVIOUS THERAPY	INVOLVEMENT	LOBECTOMY DATE	POSTOPERATIVE COURSE	PRESENT STATUS
M. M. 31 F	II, 5 years Pneumothorax, pneumonolysis, phrenic surgery	Stenosis of RUL bronchus with atelectasis; recurrent pneumonitis	RUL 9-14-46	Thoracoplasty in May 1947 for basal spread	February 1948 gastric culture "negative"; starting exercise in sanatorium; condition excellent
V. T. 27 M	III, 3 years Pneumothorax, pneumonolysis, phrenic surgery	Bronchitis right stem bronchus; recurrent pneumonitis, cavities in LL	RM and LL phrenic 8-16-46	Continued improvement	9-7-47 "negative" sputum, slight dyspnea on extreme exertion; "negative" sputum, February, 1948
L. M. 26 F	II, 5 years Phrenic surgery, extrapleural pneumothorax	Stenosis RUL bronchus, atelectasis UL; ulcerative bronchitis responded to streptomycin	RUL 10-7-47 Thoracoplasty 10-30-47	Tuberculous laryngitis; streptomycin 12-29-47 to 3-2-48 with great improvement	4-15-48 Repeated sputum cultures "negative"

destruction of lung parenchyma. One patient (B.P.) had a large mixed abscess. These 3 had pneumonectomy followed by thoracoplasty.

Tension cavity and mistaken diagnosis: Only one patient had a true tension cavity (table 10). An upper lobe lobectomy was done without subsequent thoracoplasty in this patient who refused to consider any type of surgery with resultant chest deformity. There were three instances (3.75 per cent) in which

TABLE 5

Bronchial stenosis (pneumonectomy)

PATIENT, AGE, SEX	STAGE DURATION PREVIOUS THERAPY	INVOLVEMENT	PNEUMONECTOMY DATE	POSTOPERATIVE COURSE	PRESENT STATUS
P. S. 18 F	III, 4 years. Sanatorium 4 years, phrenic surgery	Fibroid lesion, atelectasis, pinpoint stenosis left stem bronchus	Left 3-13-46	Rapid convalescence	December 1947 "negative" sputum, at home, attending school
R. B. 22 F	III, 2 years. Pneumothorax	Inexpansile atelectatic lung, high grade stenosis left stem bronchus	Left 12-26-46 Thoracoplasty 1-20-47	Uneventful	9-1-47 "negative" sputum, in sanatorium, 4-20-48 at home
R. S. 29 F	III, 7 years. Pneumothorax, phrenic surgery	Bronchial stenosis, repeated hemoptysis caseous lesions throughout	Left 3-17-47 Thoracoplasty 4-9-47	Chest films continue to remain unchanged	3-4-48 "negative" sputum, at home
R. L. 19 F	III, 4 years. Streptomycin for 4 months in sanatorium	Stenosis of left stem bronchus	Left 3-18-47 Thoracoplasty 4-2-47	Excellent convalescence	3-9-48 "negative" sputum, at home
D. K. 18 F	III, 4 years. Pneumothorax, pneumonolysis, phrenic surgery	Cavitation LUL, stenosis of stem bronchus, recurrent pneumonitis	Left 3-31-47 Thoracoplasty 4-19-47	Continued improvement	4-15-48 "negative" sputum
G. H. 44 M	II, 12 years. Pneumothorax	Stenosis of stem bronchus with widely disseminated disease, cavity UL	Left 5-15-47 Thoracoplasty 6-10-47	Streptomycin 7-24-47 to 9-7-47 because of questionable pericarditis; opposite lung stable	3-15-48 "negative" sputum
A. K. 23 F	III, 6 years. Pneumothorax, phrenic surgery; streptomycin for 90 days in sanatorium 1947	High grade stenosis stem bronchus, scattered tubercles in UL	Left 5-28-47 Thoracoplasty 6-18-47	Satisfactory convalescence	2-11-48 "negative" sputum, in sanatorium, 4-15-48 at home
J. R. 33 F	III, 6 years. Pneumoperitoneum streptomycin, pneumothorax, 2 phrenic operations	Stenosis of stem bronchus	Right 7-29-47 Thoracoplasty 8-20-47	General condition continues to improve	2-1-48 "negative" sputum
K. P. 28 M	III, 14 years. Sanatorium	Stenosis of stem bronchus with complete atelectasis	Left 8-1-47 Thoracoplasty 8-28-47	Continued improvement in sanatorium	2-1-48 "negative" sputum
A.K. 47 M	II, 1 year Streptomycin 4 months in sanatorium, pneumothorax	Atelectatic left lung with cavity, high grade stenosis	Left 9-29-47	Died of cerebral thrombosis 10-1-47	Dead
W. O. 30 M	III, 5 years. Pneumothorax, pneumonolysis, 2 phrenic operations	Stenosis of RLL bronchus, hemoptysis, bronchiectasis, severe cough	Right 11-10-47 Thoracoplasty 12-3-47	Severe dyspnea for 1 month	Left lung remains stable, 4-15-48 "negative" sputum

TABLE 5—*Concluded*

PATIENT, AGE, SEX	STAGE DURATION PREVIOUS THERAPY	INVOLVEMENT	PNEUMONECTOMY DATE	POSTOPERATIVE COURSE	PRESENT STATUS
A. H. 29 F	III, 18 years Pneumothorax, thoracotomy for mixed infection empyema	Atelectatic lung, complete bronchial stenosis, numerous cavities, bronchiectasis	Left 12-10-47 Thoracoplasty 12-30-47	Streptomycin started 2-21-48 because of reopening of old chest sinus	4-15-48 sputum cultures "negative"
B. B. 21 F	III, 2 years. Pneumoperitoneum, phrenic surgery	50 per cent stenosis of stem bronchus, active bronchitis with no ulceration	Left, with thoracoplasty 1-13-48	Opposite lung has remained stable	4-15-48 "negative" sputum

TABLE 6

Tuberculous bronchiectasis (lobectomy)

PATIENT, AGE, SEX	STAGE DURATION PREVIOUS THERAPY	INVOLVEMENT	OPERATION DATE	POSTOPERATIVE COURSE	PRESENT STATUS
H. M. 26 M	II, 7 years. Pneumothorax, phrenic surgery	Bronchiectasis, repeated hemoptysis, cavity LUL	LU lobectomy 2-19-45	Working full time at present, lesion stable by roentgenography	8-1-47 "negative" sputum, full time work, 4-15-48 "negative" sputum
B. H. 59 M	III, 3 years. Bed-rest	Bronchiectasis RU and ML, severe	RU and ML lobectomy 2-16-46	Developed bronchial fistula, putrid empyema	Died 55th postoperative day
B. K. 26 F	II, 6 months. Phrenic surgery, streptomycin 35 days in sanatorium	Bronchiectasis RU and ML, caseous lesions bronchial distribution	RU and ML lobectomy and thoracoplasty 11-12-47	Streptomycin continued until 1-18-48, total of 90 days, chest roentgenogram reveals stable lesions	February 1948 "negative" sputum
P. O. 28 F	II, 2 years Pneumothorax, phrenic surgery	Bronchiectasis, atelectasis RUL	RU lobectomy Thoracoplasty 4-23-47	Roentgenogram reveals stable lesions, streptomycin 8-23-47 to 11-25-47, no source evident	9-1-47 "positive" sputum, 1-29-48 "positive" sputum
M. M. 20 F	III, 4 years. Pneumothorax, phrenic surgery	Bronchiectasis RUL, copious sputum, contralateral pneumothorax	RU lobectomy 8-21-47. Segmental of apex LL. Thoracoplasty 9-13-47	Small bronchopleural fistula with mixed infection, closed with thoracoplasty	10-21-47 "negative" sputum, January 1948 "negative" sputum; condition excellent
M. R. 26 F	II, 9 years. Pneumothorax	Bronchiectasis LUL	LU lobectomy 8-25-47 Phrenic surgery 8-30-47 Thoracoplasty 9-11-47	Continued good during postoperative period	10-21-47 "negative" sputum, January 1948 "negative" sputum; chest stable
S. B. 20 M	III, 2 years. 2 phrenic operations	Bronchiectasis with cavity RUL	RU lobectomy 9-10-47 Thoracoplasty 10-3-47	Temporary atelectasis LL	10-1-47 "negative" sputum, 12-1-47 "negative" sputum; being rehabilitated
H. G. 36 F	III, 1 year. Streptomycin 92 days	Saccular bronchiectasis RUL with cavity	RU lobectomy and thoracoplasty 2-13-48	Uneventful	4-20-48 "negative" sputum

TABLE 7

Lower lobe lesions

PATIENT AGE, SEX	STAGE DURATION PREVIOUS THERAPY	INVOLVEMENT	OPERATION DATE	POSTOPERATIVE COURSE	PRESENT STATUS
M. S. 26 F	III, 2 years. Phrenic surgery	Cavitation RLL, copious sputum, atelectasis RM and LL	RM and LL lobectomy with phrenic surgery 11-7-46	Continued improvement	9-1-47 "negative" sputum; at home; 2-8-48 "negative" sputum; lesion stable by roentgenography.
J. DeS. 22 M	II, 5 years. Rest, pneumothorax unsatisfactory because of adhesions	Tuberculoma LLL, bronchiectasis UL	Pneumonectomy, Lt 4-23-47 Thoracoplasty 6-19-47	Continued improvement since September 1947	9-1-47 "positive" sputum in sanatorium; six "negative" cultures since November, 1947
K. K. 25 M	III, 4 years. Pneumothorax, streptomycin 81 days	Multiple cavities RLL	RLL lobectomy with phrenic surgery 9-20-47	Uneventful, continued improvement in general condition	10-4-47 "negative" sputum; 12-29-47 "negative" sputum
D. F. 17 M	III, 5 months. Rest, streptomycin 45 days	RM and LL atelectasis and cavitation	RM and LL lobectomy with phrenic surgery 11-20-47	Uneventful	2-26-48 "negative" sputum; condition excellent

TABLE 8

Pneumothorax failure

PATIENT, AGE, SEX	STAGE DURATION PREVIOUS THERAPY	INVOLVEMENT	OPERATION DATE	POSTOPERATIVE COURSE	PRESENT STATUS
M. G. 28 M	III, 6 years. Pneumothorax	Nonexpansile left lung with cavity UL	Pneumonectomy, left 8-8-47 Thoracoplasty 8-29-47	Contralateral pleural fluid became purulent with bronchopleural fistula 2-21-48	2-1-48 "positive" sputum; not good
R. M. 42 M	II, 5 years. Pneumothorax, phrenic surgery	RUL cavity failed to close with pneumothorax	RU lobectomy 9-6-47; Thoracoplasty 9-25-47	Wound infection with osteomyelitis of ribs following thoracoplasty, reoperation necessary	4-15-48 "negative" sputum
R. N. 32 M	III, 2 years. Pneumothorax pneumonolysis	Nonexpansile left lung with tuberculous empyema	LU lobectomy with decortication of LL 11-3-47; Thoracoplasty 11-17-47	Uneventful	4-15-48 "negative" sputum
C. S. 33 M	III, 1 year. Sanatorium	Spontaneous pneumothorax, inexpansile lung, tuberculous empyema	Pneumonectomy, left 1-14-48 Thoracoplasty 2-9-48 2-25-48	Uneventful	4-15-48 smears of sputum "negative"

the diagnosis of tuberculosis was overlooked previous to operation. In two older individuals the preoperative diagnosis was bronchogenic neoplasm and in one

TABLE 9

Destroyed lung (pneumonectomy)

PATIENT, AGE, SEX	STAGE DURATION PREVIOUS THERAPY	INVOLVEMENT	PNEUMONECTOMY DATE	POSTOPERATIVE COURSE	PRESENT STATUS
B. P. 30 F	III, 8 years Unsuccessful pneumothorax	Multiple cavities throughout left lung, extensive fibrosis	Left 4-25-47 Thoracoplasty 5-22-47	Atelectasis RUL, cleared	9-1-47 "negative" sputum; gained 30 lbs; 4-3-48 clinically well, at home
F. DeS. 21 F	III, 4 years Phrenic surgery, repeated 1 year later	Cavity RUL, multiple bronchiectatic cavities in lower lobes, ulcerative bronchitis right 10-20-47, red bronchus following streptomycin 11-17-47	Right 11-26-47 Thoracoplasty 12-16-47	Streptomycin started 3-28-48 in sanatorium	4-15-48 "positive" sputum; bronchial ulceration
V. J. 22 F	III, 1 year Pneumothorax, streptomycin 3 months	Atelectatic lung with scattered cavities, infection of left stem bronchus	Left 12-29-47 Thoracoplasty 1-27-48	Continues to improve	4-15-48 "negative" sputum

TABLE 10

Tension cavity (mistaken diagnosis)

PATIENT, AGE, SEX	STAGE DURATION PREVIOUS THERAPY	INVOLVEMENT	OPERATION DATE	POSTOPERATIVE COURSE	PRESENT STATUS
R. G. 28 M	III, 2 years Bed-rest	Tension cavity LUL, refused thoracoplasty	LU Lobectomy 4-10-47	Continued improvement	January 1948 "negative" sputum, playing professional baseball
J. B. 65 M	1 year No treatment	Hilar mass on right, 8 by 8 cm.; bronchoscopy negative	Pneumonectomy, right 10-1-47	Mistaken diagnosis, thought to be cancer, proved to be caseous tuberculosis	Excellent; full time work
L. E. 27 F	III, 1 month	Bronchiectasis, atelectasis of RUL, stenosis of UL bronchus	RU Lobectomy and thoracoplasty 10-11-47	Thought to be a nontuberculous bronchiectasis; sputum "negative" before operation; histopathologic examination revealed tuberculosis	4-15-48 "negative" sputum; excellent
D. H. 59 F	4 to 5 years Deaf mute	Obstruction in LLL bronchus	Pneumonectomy, left 11-18-47 Followed by auricular fibril., cardiac decomp.; digitalis	Thought to be a bronchiogenic cancer before operation	4-15-48 "negative" sputum

other the preoperative diagnosis was nontuberculous bronchiectasis secondary to pneumonia. In all of these patients the roentgenographic appearance was not characteristic of tuberculosis. In addition to these findings, several sputum

smears and cultures examined preoperatively were negative for tubercle bacilli. However, the pathologic picture of the resected tissue was characteristic of tuberculosis.

In table 11 may be seen the number of patients having pneumonectomy and lobectomy under the different indications chosen for resection. The greatest number (41) were performed for thoracoplasty failure. This is not surprising as most of the resections fell under this indication. Forty-seven (58.75 per cent) of the patients had pneumonectomy and 33 (41.25 per cent) had lobectomy.

TABLE 11

Type of operation

INDICATIONS	PNEUMONECTOMY	LOBECTOMY	OPERATIVE DEATHS
Thoracoplasty failure	26	15	—
Bronchial stenosis	13	3	1 (Pneumonectomy)
Tuberculous bronchiectasis	—	8	1 (Lobectomy)
Lower lobe lesions	1	3	—
Pneumothorax failure	2	2	—
Destroyed lung	3	—	—
Mistaken diagnosis	2	1	—
Tension cavity	—	1	—
Total 80 patients	47 (58.75 per cent)	33 (41.25 per cent)	Operative mortality 2.5 per cent

RESULTS

In the entire group of 80 resections there were 2 operative deaths, giving a total series mortality of 2.5 per cent. One patient (B. H.) died on the fifty-fifth postoperative day with a bronchopleural fistula and a mixed empyema. The other (A. K.) died on the first postoperative day. Postmortem revealed the cause of death to be thrombosis of a cerebral artery.

The various postoperative complications are listed in table 12. Some were of a temporary nature but are included to make the report complete and self-explanatory.

The results in relation to indications are presented in table 13. In view of the short period of observation which has passed since many of the resections, it is not surprising that the 52.5 per cent of the total group, who are apparently noninfectious, still remain in the sanatorium. Only 25 patients have been discharged. The total of 83.75 per cent of the series with cultures negative for tubercle bacilli exceeded expectations. A high percentage of reversal of infectiousness occurred in the cases from the three groups representing the most common indications for the procedure.

In table 14 may be seen the final results obtained in the series of 80 patients, as well as the number of reactivations or "spreads" in the ipso-and contralateral lung. There were 47 pneumonectomies and 33 lobectomies. Each procedure was complicated with four "spreads" or reactivations. No definite conclusions

can be drawn, however, from such a small and recently operated group of cases. Reversal of infectiousness occurred in 83.75 per cent of the entire group. Three

TABLE 12

Complications following pneumonectomy and lobectomy

	PNEUMONECTOMY (47 CASES)	LOBECTOMY (33 CASES)
Spread		
Ipsolateral	—	2
Contralateral	4	2
Dyspnea	1 (for 4 weeks)	1 (temporary)
Reactivation of other tuberculous foci	—	1 (middle ear) 1 (larynx)
Sinus of chest wall (tuberculous)	2	—
Ulceration of bronchial stump	2	—
Bronchopleural fistula	—	1 (with empyema, died) 1 (temporary)
Pleural effusion	2 (contralateral)	—
Wound infection	—	1 (osteomyelitis of rib stumps)
Hemothorax	1	—
Streptomycin reaction	1	1
Respiratory cripple	1	—

TABLE 13

Results in relation to indications (pneumonectomy and lobectomy)

	NUMBER	NEGATIVE		POSITIVE		DEAD
		Sanatorium	Home	Sanatorium	Home	
Thoracoplasty failure	41	21	12	5	—	3
Stenosis of bronchus	16	9	6	—	—	1
Bronchiectasis	8	5	1	1	—	1
Lower lobe lesions	4	3	1	—	—	—
Pneumothorax failure	4	3	—	1	—	—
Destroyed lung	3	1	1	1	—	—
Mistaken diagnosis	3	—	3	—	—	—
Tension cavity	1	—	1	—	—	—
Total	80 (100 per cent)	42 (52.5 per cent)	25 (31.25 per cent)	8 (10 per cent)	—	5 (6.25 per cent)

patients have died since leaving the hospital, but in only one of these was death due to tuberculosis. Two deaths occurred in older individuals. One was prob-

ably due to right heart failure while the other, which was sudden, was presumably the result of a myocardial infarction.

The duration of the state of noninfectiousness since operation is listed in table 15. As may be seen, 52 patients (65 per cent) are recorded in the "less than eighteen months" period. Fifteen (18.75 per cent) have remained noninfectious over periods of eighteen months to three years. As only 3 of the living patients

TABLE 14

Results of resection (all patients)

	NUMBER	REACTIVATION OR SPREAD		EXAMINATION OF SPUTUM FOR TUBERCLE BACILLI		DEAD
		Ipso-lateral	Contra-lateral	Positive	Negative	
Pneumonectomy						
Right	18	—	2	2	15	1
Left	29	—	2	3	24	2
Lobectomy						
One lobe	21	2	2	2	18	1
Two lobes	11	—	—	1	9	1
Segmental	1	—	—	—	1	—
Total	80 (100 per cent)			8 (10 per cent)	67 (83.75 per cent)	5 (6.25 per cent)

TABLE 15

Duration of conversions since operation

	1 to 6 MONTHS	6 to 12 MONTHS	12 to 18 MONTHS	18 to 24 MONTHS	24 to 30 MONTHS	30 to 36 MONTHS	36 MONTHS
Thoracoplasty failure	7	7	6	6	1	4	1
Bronchostenosis	3	5	6	1	1	—	—
Bronchiectasis	2	3	—	—	—	—	1
Lower lobe lesion	1	1	2	—	—	—	—
Pneumothorax failure	1	2	—	—	—	—	—
Destroyed lung	1	—	1	—	—	—	—
Mistaken diagnosis	1	2	—	—	—	—	—
Tension cavity	—	—	1	—	—	—	—
	16	20	16	7	2	4	2

have shown "spread" or reactivation, it appears that not many complications should be expected in this group in the future. It is realized, however, that tuberculosis is an unpredictable disease and as time passes some of these apparently excellent results may eventually have to be classified as failures.

Streptomycin therapy prior to operation has been used in 26 of the 80 patients included in this group of pulmonary resections. The length of preoperative therapy has varied from four to one hundred and eighty days, but only 10 patients have had streptomycin for more than thirty-one days before surgery.

In these cases the indication for treatment was tuberculous endobronchitis. The control of their acute symptoms and the recognition of bronchostenosis led to the selection of pulmonary resection. In only 2 patients did the persistence of tuberculous bronchitis necessitate the prolongation of streptomycin for ninety to one hundred and thirty-six days. Three patients treated early in 1947 received 3.0 Gm. of streptomycin per day, one patient 2.0 Gm. per day, and three 1.5 Gm. per day. The remaining 19 patients received 1.0 Gm. per day.

With few exceptions, the 16 patients who were treated for less than thirty-one days were likewise treated because of bronchial disease. The bronchoscopic examination revealed bronchitis in most of these individuals, though there were several with upper lobe disease in which bronchial involvement, presumably tuberculous, was felt to be present from the findings on physical and roentgenologic examinations. Three patients with tuberculous empyema were treated for short periods preoperatively.

Streptomycin by the intramuscular route has been used postoperatively in 37 patients. The average length of treatment was for fourteen days with the usual dose of 1.0 Gm. per day.

From limited experience with streptomycin, the writers believe that it is of great value in the control of acute tuberculous bronchitis. Before the use of antibiotics these cases either had to be denied surgery or be operated upon at a considerably increased risk. A fair number of the group included under thoracoplasty failure had acute tuberculous bronchitis at the time of thoracoplasty. The bronchial lesion subsequently healed so that resection could be done.

In addition to the parenteral use of streptomycin in the postoperative period, the majority of the patients have received streptomycin as well as penicillin locally. The use of these drugs in lobectomies, where there must be constant postoperative closed drainage, has been limited to instillation in the wound at surgery. In postoperative pneumonectomies streptomycin and penicillin have been introduced into the pleural cavity at the time of aspirations until the fluid was well controlled or the space obliterated by thoracoplasty. The average length of such therapy was approximately ten days.

Experience with the postoperative group has been much too recent to attempt evaluation. Whether there will be any fewer late postoperative "spreads" or reactivation remains to be determined. In only one patient was it believed that a small contralateral "spread" occurred, although the lesion may have been atelectasis. This patient received streptomycin for twenty-six days and her course in the past nine months has been satisfactory. The gastric contents six weeks after surgery were negative for tubercle bacilli on culture.

The ideal length of postoperative streptomycin therapy is undetermined. The short period of therapy used in this series was selected arbitrarily. However, such a period of therapy is well worth evaluation. If "spill" to the other portion of the lung or lungs occurs, as seems likely, streptomycin should have an extremely beneficial effect. With this short period of treatment the phenomenon of streptomycin-resistance is less frequently encountered so that later spreads or reactivation might be benefitted by further treatment.

DISCUSSION

The choice of resection as a form of therapy came after several years of experience with other forms of collapse therapy. During the past ten years the writers have seen an increasing number of thoracoplasty failures and careful evaluation of these demonstrated that there were definite reasons for the failures. At times limited rib resection resulted in incomplete collapse after the first stage. Many of these were still ineffective after extensive revisions. Improperly selected cases were found to have been subjected to thoracoplasty before it was realized that the lesions were not suitable for this form of therapy. Large tension cavities at the apex, thick-walled cavities close to the mediastinum, high grade stenosis of the bronchus (often lobar) and bronchiectasis accounted for the major group of failures. The dictum that thoracoplasty should always be given first trial continued to be followed. Most patients showed improvement but the goal, that is, the elimination of tubercle bacilli, was not reached. The number of failures that accumulated as time passed, coupled with the newly developed surgical technique of individual ligation of the hilar structures with its associated low mortality, stimulated the writers to try to accomplish something for this discouraged group of patients. The availability of penicillin and streptomycin permitted operation on those patients who would have been refused for lack of a means of controlling ulcerative bronchitis. It now seems evident that thoracoplasty as previously used served only as a stopgap in the treatment of these cases.

The most frequent indication for resection in this series was thoracoplasty failure (41 cases). The next important indication was high grade bronchostenosis (16 cases). Various complications were also present in the form of hemoptysis, recurrent attacks of pneumonitis, sepsis and atelectasis. It was in the second group that the removal of the involved pulmonary tissue produced striking early results. The results in these two groups of cases have been most gratifying to date.

The remaining 30 per cent of the series presented individual problems as shown in the various tables. Most of these had received various therapeutic procedures before being considered for resection. A few cases were chosen for resection as the primary procedure because experience indicated that any other method would most likely fail.

Thoracoplasty was performed 26 times following resection. It is believed that this is advisable in all cases except those with lower lobe resection. In the more recent cases thoracoplasty was performed at the time of the resection if the patient's condition permitted, or as soon thereafter as possible. Following lobectomy, thoracoplasty ensures against over-expansion of the remaining lobe or lobes and should protect against ipsolateral reactivation. Thoracoplasty is equally important following pneumonectomy as it prevents a shift of the mediastinum with accompanying over-expansion of the contralateral lung and promotes early stabilization. Not infrequently tuberculous tissue is cut through during the operation and a thoracoplasty extensive enough to obliterate the dead space completely appears necessary if tuberculous empyema is to be avoided.

In several recent cases pneumonectomy has been performed in the presence of a tuberculous or mixed empyema and the infection controlled with frequent aspirations and the instillation of penicillin and streptomycin until a sufficiently adequate thoracoplasty could be done to obliterate the pleural space.

Phrenicotomy was originally performed following upper lobe lobectomy with the thought that reduction in the size of the pleural space would permit the remaining lobe to fill the space more quickly. It is now believed that phrenicotomy reduces the efficiency of the cough and promotes atelectasis. Thoracoplasty is done instead unless the lower lobe has been removed. In this instance phrenic section is used routinely as a substitute for thoracoplasty.

SUMMARY

Experience with 80 consecutive cases of pulmonary resection for tuberculosis is reported. The majority of these patients were failures with the usual collapse procedures and were doomed to continued sanatorium care because of persistent infectiousness.

Sixty-seven cases (83.75 per cent) of the entire group are noninfectious, 8 (10 per cent) still discharge tubercle bacilli and 5 (6.25 per cent) are dead. There were 2 operative deaths, yielding an operative mortality of 2.5 per cent.

A sufficient length of time has not passed to draw definite conclusions regarding the permanency of these results. If the cases follow the course reported by other authors, the satisfactory results will diminish with time. It must not be forgotten that tuberculosis is a systemic disease with many vagaries and that the removal of the symptom-producing focus is not the complete solution of the problem. Time has also shown that collapse therapy in the form of thoracoplasty has been found wanting under certain circumstances. Late spread and reactivation following thoracoplasty or resection should not be cited to condemn either procedure. Further experience with the methods at hand and newer methods may supply the answer.

Streptomycin when available was used before and after operation. It has proved most valuable in cases with acute ulcerative bronchitis. Many of the resections in the present series would have been ill advised and dangerous before streptomycin was available. The drug is given in doses of 1.0 Gm. daily. Frequent instillations into the pleural cavity following pneumonectomy have prevented the common complication of tuberculous empyema until the space was obliterated. It is believed that chemotherapy is partially responsible for the low number of reactivations and "spreads" and that it has helped to control them once they have occurred. Penicillin has been used extensively throughout the series.

SUMARIO

La Resección Pulmonar en el Tratamiento de la Tuberculosis

Comunícase lo observado en 80 casos consecutivos de resección pulmonar por tuberculosis. La mayoría de los enfermos representaban fracasos de los habituales procedimientos de colapso y estaban condenados a continua asistencia sanatorial debido a su infecciosidad persistente.

Sesenta y siete casos (83.75 por ciento) del grupo total ya no son infecciosos, 8 (10 por ciento) todavía expulsan bacilos tuberculosos y 5 (6.25 por ciento) han muerto. Hubo 2 muertes quirúrgicas, dando una mortalidad operatoria de 2.5 por ciento.

No ha trascurrido aun suficiente tiempo para poder sacar conclusiones definitivas acerca de la permanencia de los resultados. Si los casos se conforman al patrón presentado por otros autores, los resultados satisfactorios irán disminuyendo con el tiempo. No hay que olvidar que la tuberculosis es una enfermedad orgánica y bastante caprichosa y que la eliminación del foco sintomatógeno no representa la solución completa del problema. El tiempo también ha revelado que la colapsoterapia en forma de toracoplastia ha resultado deficiente en ciertas circunstancias. No deben citarse la difusión y reactivación tardias después de la toracoplastia o la resección para condenar uno u otro procedimiento. Nuevas tentativas con las técnicas disponibles y con otras más nuevas tal vez faciliten la respuesta.

La estreptomicina, cuando la había, fué utilizada antes y después de la intervención, resultando más valiosa en los casos con bronquitis ulcerada aguda. En la serie actual, muchas de las resecciones hubieran sido indiscretas y peligrosas antes de contarse con la estreptomicina. La droga fué administrada a dosis de 1 Gm. diario. Las frecuentes instilaciones en la cavidad pleural consecutivamente a la neumectomía han impedido las habituales complicaciones del empiema tuberculoso hasta la obliteración del espacio. La quimioterapia parece ser en parte la causa del pequeño número de reactivaciones y "difusiones" y también parece haber ayudado a cohibirlas, una vez aparecidas. La penicilina fué empleada en gran escala en toda la serie.

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DECORTICATION OF THE LUNG IN PATIENTS WITH PULMONARY TUBERCULOSIS[1, 2]

PAUL V. O'ROURKE, E. J. O'BRIEN, AND WM. L. TUTTLE

INTRODUCTION

The results during World War II of decortication of the lung in the management of organizing hemothorax are well known and accepted. Some thoracic surgeons hoped that this technique could be applied successfully in certain tuberculous patients with a persistent pleural space in whom conservative measures had failed. This hope was based on the belief that the thickened pleura seen in those cases of tuberculosis was in reality a fairly normal visceral pleura covered by an organized exudate or "peel" similar in many ways to that seen in organizing hemothorax. Fear had been expressed that the "peel" could not be removed from the visceral pleura because of its firm adherence; that the complications occurring from its removal, if possible, would be very great; and that tuberculous wound infections and failure to obliterate the pleural space would occur frequently enough to exclude its common usage.

This study was undertaken by the writers who are fully aware of these possibilities but anxious to learn if decortication is surgically feasible in tuberculosis. It consists of 43 cases of pulmonary and pleural tuberculosis decorticated between January 1946 and May 1948. There were 30 males and 13 females; 38 were white and 5 were Negroes.

INDICATIONS FOR DECORTICATION

For the most part only patients with complications of tuberculous pleurisy with effusion or pneumothorax were considered for this operation. It was used also as an adjunct to lobectomy when the need arose. The indications for decortication in the group having tuberculous pleurisy with effusion have been restricted to those patients in whom: (a) the fluid became organized or loculated and could no longer be removed by aspiration; or (b) the fluid had been aspirated and replaced by air resulting in an unexpandable lung. As shown in table 1, there were 9 cases in this group. In 2 of the 5 patients with loculated fluid which failed to respond to aspiration a positive diagnosis of pleural tuberculosis was not made preoperatively, but was made histologically from the "peel." Of the 2 patients with tuberculous empyema, one occurred in the youngest patient, four years old, who had been a contact of his mother before she was discovered to have tuberculosis (figure 1). In the patient with a mixed empyema the process began as a tuberculous pleural effusion, which was aspirated for two years with little benefit. After a routine aspiration, the patient developed fever and the

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[2] Presented before the Medical Section, as part of the *Symposium on Surgery—Trends—Late Results*, at the 44th Annual Meeting of the National Tuberculosis Association, New York, New York, June 16, 1948.

TABLE 1

Indications for decortication

	NUMBER OF PATIENTS	
I. Complications of Tuberculous Pleurisy		
a. Unexpandable lung	1	
b. Mixed empyema with unexpandable lung	1	
c. Tuberculous empyema with unexpandable lung	2	
d. Loculated fluid with unexpandable lung	5	
Total		9
II. Complications of Pneumothorax		
a. Unexpandable lung	3	
b. Mixed empyema with unexpandable lung	3	
c. Tuberculous empyema with unexpandable lung	7	
d. Pleural effusion with unexpandable lung	14	
Total		27
III. Adjunct to lobectomy	7	7
Total		43

FIG. 1a. (Left) Preoperative film dated March 6, 1948. Age 4 years. Tuberculous pleurisy with empyema.

FIG. 1b. (Right) Postoperative film dated May 10, 1948. Decortication done March 15, 1948.

fluid, which had always been clear, became purulent. *Staphylococcus aureus* was recovered upon culture of the fluid from the pleural cavity. The infection was immediately drained. One year after the drainage, because of the persistent

pleural space, a decortication was done successfully. Re-expansion of the lung at the end of the operation was good but postoperatively the expansion was not sustained.

The second indication used by the writers have been the complications which accompany or follow therapeutic pneumothorax, such as troublesome fluid or empyema occurring during the program of pneumothorax, or failure of the lung to re-expand after pneumothorax had been abandoned. There were 27 patients in this group. Ten patients had tuberculous or mixed empyema which failed to respond to pleural lavage and the lung remained collapsed after aspiration of air or fluid. There were 14 patients with pleural effusion occurring during the re-expansion program or during the period of the therapeutic pneumothorax, in which the lung failed to re-expand after aspiration and discontinuance of air refills. This is the largest single group for which a decortication has been performed. The encased or unexpandable lung without fluid has been decorticated in three instances to obliterate the pleural space and to prevent shift of the mediastinum and over-distention of the contralateral lung. In the beginning this particular indication was used hesitantly because most of these patients were at home and apparently doing well. It seemed inadvisable to change an apparently satisfactory program until it was apparent that decortication could be done with relative safety. In 2 of the 3 cases in this group the indications for decortications were impelling. One, a woman who had been receiving pneumothorax refills for six years, had some bleeding into the pleural space with the formation of a large clot following a refill of air. The pleura above the clot showed thickening on the roentgenogram and re-expansion of the lung seemed unlikely unless a decortication could be done. After the clot had been removed the lung would not re-expand even though positive pressure was applied through an intratracheal tube. A decortication was done easily and complete re-expansion was obtained (figure 2). The second patient was a doctor engaged in tuberculosis work who had been observing the results of the study and requested a decortication of his unexpanded lung. His wishes were finally granted and an excellent result was obtained. As the encased lung without fluid seems to represent one of the better indications, it is planned to use it with increasing frequency in the future.

The third indication is represented by a group of 7 patients upon whom a lobectomy was performed and a "peel" removed from the remaining lobes to aid in their re-expansion and to hasten obliteration of the pleural space. Incidentally, in 3 pneumonectomies not included in this series a decortication was first done to facilitate the indentification and handling of the hilar structures.

The duration of the pneumothorax, as listed in table 2, has varied from four months to sixteen years and in five instances has been present for ten years. Ability to separate the "peel" from the visceral pleura does not seem to be related to the duration of the pneumothorax *per se* but rather to the amount and extent of the peripheral pulmonary disease.

Unless some emergency arose requiring surgical intervention, an adequate trial of re-expansion was attempted and only when further efforts seemed hazardous or futile was decortication employed.

A complete decortication was not always possible or desirable, because of the underlying disease, nor always necessary because of the small size of the remaining

FIG. 2a (Left) Preoperative film dated September 18, 1947. Duration of pneumothorax six years. Bleeding and formation of clot following refill.

FIG. 2b (Right) Postoperative film dated April 19, 1948. Decortication done September 22, 1947.

TABLE 2

Duration of pneumothorax

	NUMBER OF PATIENTS
Less than one year	4
Two years	5
Three years	4
Five years	3
Six years	2
Seven years	1
Eight years	1
Ten years	5
Fourteen years	1
Sixteen years	1
Total	27

pleural space. Twenty-eight lungs were completely decorticated and 15 partially. Early in the study it was observed that a lobe containing considerable fibrosis as a result of previous disease did not re-expand well even though it was completely decorticated. Since then, if the disease-free lobes after decortication

filled the hemithorax, no attempt has been made to remove the "peel" from the lobe which was the site of the major disease. Perhaps it is better to keep the diseased lobe permanently collapsed than risk the danger of reactivation from re-expansion.

The majority of patients in this series were subjected to the operation without streptomycin as an adjunct because it was not available. More recently, however, streptomycin has been used in 19 instances either because of complications encountered at operation, such as subpleural tubercles, or to prepare the patient for the procedure. It is believed at present that practically all individuals should receive streptomycin as a routine procedure if for no other reason than prophylaxis. It should be emphasized, however, that the paramount consideration for success is dependent upon adequate removal of the "peel" without undue trauma to the underlying lung. That streptomycin may improve the result and hasten the convalescence seems indicated but it is no substitute for good surgery.

TECHNIQUE OF OPERATION

The actual technique of the operation did not differ from that used by thoracic surgeons during World War II in the treatment of organizing hemothorax. All operations were done with intratracheal anesthesia of nitrous oxide-oxygen and ether. Approximately 1,500 cc. of blood were given during the procedure. Cotton was used for ligatures and wound closure. In most instances one medium sized rubber tube drain was placed dependently just above the diaphragm in the posterior axillary line and a smaller tube was inserted high anteriorly to allow for the escape of air. Both tubes were connected to water-sealed bottles.

The immediate postoperative course did not differ from that of the usual patient with tuberculosis undergoing surgery. Fever was not excessive or prolonged except in the one patient who died three months after operation from disseminated tuberculosis. Two patients needed bronchoscopy to remove retained bronchial secretions.

As soon as the amount of drainage stopped fluctuating, the tubes were removed. The tubes were withdrawn, on an average, by the fifth day in 31 cases. If fluoroscopy or roentgenograms showed the presence of loculated fluid or trapped air, they were removed by aspiration. Nine patients required frequent thoracentesis over a period of several weeks. During the time of re-expansion they were watched very closely and every effort was made to keep the pleural space dry and as free from trapped air as possible. Deep breathing exercises were started as soon as the tubes were removed and were continued indefinitely.

COMPLICATIONS

In 5 patients there were seven complications attributable to the operation, as shown in table 3. One patient with pleural tuberculosis and empyema did non re-expand at the completion of the operation despite the successful removal of a "peel". In 3 additional patients a partially *successful decortication* was done and the immediate re-expansion of the lung seemed good when positive

pressure was used. Complete re-expansion was not sustained, however, during the postoperative period. Two of the latter have had a thoracoplasty performed which obliterated the pleural space and they are now at home.

At the time of operation it was not infrequent that small bubbling air leaks were seen. In all but 3 instances these were not of any consequence and apparently sealed off within a few hours. Of the 3 patients in whom air leakage continued, 2 developed mixed empyema and were drained. In one patient the fistula closed soon after drainage. In the other, the fistula has remained open and the prognosis is unfavorable. The third instance of bronchial fistula was

TABLE 3

Immediate postoperative complications

	OCCURRENCE*
Expansion of lung not sustained	3
Nonexpansion of lung	1
Bronchial pleural fistula with mixed empyema	2
Bronchial pleural fistula (only)	1
Total	7

* The seven complications occurred in a total of 5 patients.

TABLE 4

Late complications of disease

	OCCURRENCE	NUMBER OF PATIENTS
Positive sputum (only)	2	2
Bilateral reactivation	1	1
Disseminated tuberculosis (death)	1	1
Incisional sinus infection only (4 months)	1	1
Total	5	5

uncomplicated by infection. In this case a posterior thoracotomy tube was converted to an open drainage tube and the space gradually obliterated and the fistula closed.

In 5 patients there were late complications, as listed in table 4. Two developed sputum positive for tubercle bacilli. In another patient a single culture positive for tubercle bacilli was obtained within one month of operation. This patient later "converted" and she has remained noninfectious. The other patient ultimately "converted" and has been noninfectious for eleven months. In neither was there roentgenological evidence of reactivation nor was other therapy except bed-rest added to influence the bacteriologic status. One patient had bilateral reactivation of disease with reopening of cavity in the ipsilateral lung. This was the only instance in the entire series of a previously closed cavity reopening after

the procedure. This patient probably contributed to his own breakdown because of leaving the hospital against advice. He was later readmitted with evident spread of his disease. One patient had widespread dissemination of disease resulting in ultimate death. This patient was a colored male who originally had bilateral disease and developed a tuberculous empyema as a complication of pneumothorax. The decortication was not difficult and re-expansion was very good at the end of the operation. The expansion was not sustained, however, and his course was one of steady progression of the disease. This resulted in death three months after operation. Streptomycin might have altered the course of his disease but it was not available when his operation was performed.

RESULTS

The results of decortication of the lung in patients with tuberculosis have been evaluated in this series by ability to obliterate the persistent pleural space. If

TABLE 5

*Results of decortication**

	NUMBER OF PATIENTS
Failure	4
Improved	5
Good	14
Excellent	20
Total	43

* In an additional 4 patients decortication could not be performed.

this objective can be obtained, it justifies the procedure in most instances because it lessens the chance of the existing pleural space becoming infected. Also it prevents emphysema developing in the contralateral lung as a result of the mediastinum shifting to aid in obliterating the pleural space. This occurs when most pneumothoraces are discontinued and an effort is made to re-expand the lung. Although patients have voluntarily stated that their breathing was improved after the operation, this may not mean that there has been any restoration of pulmonary function. Even though the pulmonary bed has been increased in size, the ability to exchange oxygen may not have been improved and the amount of unoxygenated blood may be increased.

There were 4 patients in whom expansion of the lung was not sustained and they have been classified as failures of operation.

If the pleural space was entirely obliterated and the lung appeared to be fully aerated, this has been classified as an excellent result. There were 20 patients in this category including the 7 lobectomies.

If the space was obliterated but shadows were present indicative of incomplete re-expansion of the lung, the result has been classified as good. In this group there were 14 patients.

If there was increased aeration of the lung and the pleural space was smaller, the result was listed as improved. In this group there were 5 patients.

In 4 additional patients, not included in this series, a decortication was attempted but the "peel" could not be separated from the visceral pleura and it was necessary to do a thoracoplasty.

It will be noted by referring to table 6 that of the 43 patients operated upon, 31 are well and at home. Eleven patients are still in the hospital. Four of this group are ready for discharge and 4 have been recently operated upon and are progressing satisfactorily. One patient developed a tuberculous wound sinus but this is healing. Two are resident in the hospital because of poor progress; one with a bilateral reactivation of disease and the other with a bronchial pleural fistula and mixed empyema. The ultimate outcome of the latter patient is most uncertain.

TABLE 6

Present status

	NUMBER OF PATIENTS
At home	31
In hospital	11
Dead	1
Total	43

SUMMARY

1. The lungs of 43 patients with tuberculosis have been decorticated successfully, thereby demonstrating the presence of a "peel" similar to the organized exudate found encasing the lung in organizing hemothorax. In 4 additional patients decortication was not technically feasible.

2. The indications for decortication have been restricted to the complications of tuberculous pleurisy in 9 patients and the complications of therapeutic pneumothorax in 27 patients. In addition, 7 patients were decorticated in conjunction with a lobectomy.

3. The success or failure to remove a "peel" does not seem to be related to the duration of the pneumothorax but rather to the amount of the peripheral pulmonary disease.

4. Twenty-eight lungs were completely decorticated and in fifteen a partial decortication was performed.

5. Streptomycin was not available at the beginning of this study but since becoming more plentiful has been used routinely.

6. The immediate postoperative complications were: nonexpansion of the lung, expansion not sustained, and bronchial pleural fistula with or without mixed empyema.

7. The late complications of disease were: positive sputum only, 2 patients; bilateral reactivation of disease with reopening of cavity in the decorticated

lung, one patient; tuberculous incisional sinus infection, one patient; and disseminated tuberculosis with ultimate death, one patient.

8. The results of decortication in the 43 patients have been classified as: failure, 4; improved, 5; good, 14; and excellent, 20. The effect on pulmonary function is discussed.

9. The present status of the patients is as follows: 31 at home, 11 in hospital, and one dead.

CONCLUSIONS

Decortication is a valuable adjunct to the armamentarium for the treatment of pulmonary and pleural tuberculosis. The results obtained warrant its continued usage, accompanied by further investigations of the respiratory function of the decorticated lung.

SUMARIO

La Decorticación del Pulmón en los Tuberculosos Pulmonares

1. Los pulmones de 43 tuberculosos fueron decorticados con éxito, demostrando así la presencia de una "cápsula" semejante al exudado organizado que encierra el pulmón en el hemotórax organizado. En otros 4 enfermos la decorticación no resultó técnicamente factible.

2. Las indicaciones de la decorticación fueron limitadas a las complicaciones de la pleuresía tuberculosa en 9 enfermos y a las del neumotórax terapéutico en 27. Además, 7 enfermos fueron decorticados en relación con una lobectomía.

3. El éxito o fracaso en la separación de la "capsula" no parece guardar relación con la duración del neumotórax, sino más bien con la extensión de la neumopatía periférica.

4. En 38 pulmones la decorticación fué total y en 15 parcial.

5. Al comienzo del estudio no había estreptomicina a mano, pero desde que ha sido más obtenible, se ha empleado sistemáticamente.

6. Las inmediatas complicaciones postoperatorias fueron: inexpansión del pulmón, expansión insostenida, y fístula pleurobronquial con o sin empiema mixto.

7. Las complicaciones tardías de la enfermedad fueron: solamente esputo positivo, 2 enfermos; reactivación bilateral de la dolencia con reapertura de la caverna en el pulmón decorticado, 1 enfermo; infección tuberculosa de la fístula incisional, 1 enfermo; y granulia culminando en la muerte, 1 enfermo.

8. El resultado de la decorticación en los 43 enfermos fué clasificado así: fracaso, 4; mejoría, 5; bueno, 14; y excelente, 20. También se discute el efecto sobre la función pulmonar.

9. El estado actual de los enfermos es éste: 31 en sus casas, 11 en el hospital, y uno ha muerto.

CONCLUSIONES

La decorticación constituye una valiosa adición al arsenal dedicado al tratamiento de la tuberculosis pulmonar y pleural. El resultado obtenido justifica la continuación de su empleo, junto con nuevas investigaciones de la función respiratoria del pulmón decorticado.

PULMONARY TUBERCULOSIS INVOLVING THE LOWER LOBES[1, 2]

EMIL ROTHSTEIN

INTRODUCTION

Pulmonary tuberculosis involving the lower lobes is a phenomenon of recognized occurrence and is sufficiently common to warrant consideration as a diagnostic problem in every lower lobe pulmonary lesion.

Because of its location, lower lobe tuberculosis presents definite problems in diagnosis and treatment which are different in certain ways from those encountered in upper lobe disease. In addition, there has recently arisen an immunologic problem in the pathogenesis of lower lobe disease, suggested by the work of Dr. William Dock (1). The present study was attempted in an effort to explore these problems.

Definition

In this study the following types of lesions have been observed: (*a*) all cases with the chief or only lesion immediately above the diaphragmatic shadow; (*b*) all cases with the chief or only lesion in the lower one-half of the lung fields when by appropriate lateral or oblique views these were demonstrated to lie within the anatomical boundaries of either lower lobe. Cases in which extension or spread to the lower lobe occurred from known pre-existing upper lobe disease have not been included. In each case the first roentgenogram, at least, showed the lower lobe involved and the upper lung fields essentially clear. In addition, bacteriologic proof of the tuberculous etiology has been obtained in each case.

OBSERVATIONS

Forty-eight cases of lower lobe tuberculosis have been studied from the patient population of the U. S. Veterans Administration Hospital, Wood, Wisconsin. This service is a tuberculosis unit of 430 beds. The cases have been observed for an average period of three years. No effort was made to calculate the percentage of lower lobe disease as the cases were not chosen from consecutive admissions.

Incidence: All but one of the patients were males. It is recognized (2, 3) that lower lobe tuberculosis is more common in females but there is no regular tuberculosis service for female patients at Wood. The average age of the patients was 32 years while the age of a control group was 41. This incidence of lower lobe disease in a younger age group has been found by various other investigators as well. Six infections were in Negroes.

[1] From the U. S. Veterans Administration Hospital, Tuberculosis Division, Wood, Wisconsin.

[2] Published with permission of the Chief Medical Director, Department of Medicine and Surgery, Veterans Administration, who assumes no responsibility for the opinions expressed or conclusions drawn by the author.

Diagnosis and Cause of Illness

Although lower lobe tuberculosis is fairly common, it still does not receive the attention it deserves from the standpoint of diagnosis. Of the total group, 24, or one-half, presented initial difficulties in diagnosis. This led to delayed recognition of from a few weeks or months to several years. In most delayed diagnoses in tuberculosis patients the error lies in failing to obtain chest roentgenograms, but in lower lobe disease the delay may occur despite positive roentgenologic findings. The most common error in this regard was to consider an infiltrative lesion as nontuberculous despite suggestive evidence of cavitation in some instances. Frequently this resulted in the return of the patient to duty or his discharge from the hospital while the chest roentgenograms still revealed definite disease. Because of its location, the lesion was usually considered as an unresolved or slowly resolving nontuberculous "virus" pneumonitis and was apparently thought to be of little ultimate significance. In several cases bronchoscopy and bronchography were performed because of a suspicion of bronchiectasis or new growth. In two cases neoplasm could not be excluded despite considerable study and resection was performed. Another reason for failure to diagnose lower lobe tuberculosis is the frequency of lesions, usually cavitary, which are in the apex of a lobe. On the usual postero-anterior film these lesions may be projected into the hilar shadow. This is an area where annular shadows of vascular origin are common, and small cavities here are relatively easily overlooked. In a few instances the lesion was partly or entirely hidden by the heart shadow. In one case a 4 cm. cavity was so completely obscured by the heart as to permit a postero-anterior film to be interpreted as normal except for some cardiac shift to the left. In a number of cases in which the diagnosis was overlooked, the patient returned to medical observation only after considerable extension of the disease had occurred; in several instances this was severe enough to make recovery impossible.

Extent and distribution of lesions: Of the entire group only 2 were minimal at the time of admission to Wood; 24, or 50 per cent, were moderately advanced and the remainder far advanced. This overwhelming incidence of advanced disease would not have been unusual in a prewar series of routine hospital admissions for tuberculosis. The situation has been considerably changed, however, as a result of the use of chest roentgenograms as a part of routine examinations in the service. Consequently, if every tuberculous lesion were diagnosed on the first film, it would be anticipated that the percentage of minimal lesions would be high. The reason this is not the case with lower lobe lesions may be explained by the fact that 5 cases which were minimal on the first roentgenographic examination were moderately or far advanced upon first diagnosis. Others of this series were diagnosed as "pneumonia" months before the presence of tuberculosis was recognized. In many such instances it is reasonable to assume that lower lobe physical signs, in the absence of a roentgenogram, led to an appreciable delay in arriving at the correct diagnosis.

An analysis of the anatomical distributions and characteristics of these lesions revealed that 30, or 60 per cent, were at the apex of the right or the left lower lobe

and most of these were so-called hilar cavities; the remainder were in the basal portion of the lower lobes. Twenty per cent of the lesions were predominantly infiltrative, 60 per cent cavitary and 18 per cent pneumonic. In one case the lesion could be classified as a tuberculoma. Lateral views of the "hilar" cavities were especially interesting and almost always showed the lesions to be posterior, overlying the vertebral bodies. Figures 1 and 2 show an unusually large lower lobe cavity and its posterior location.

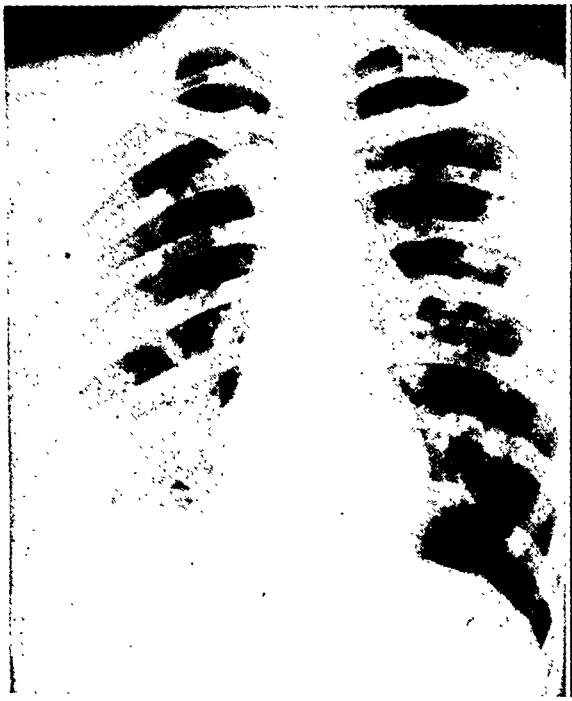

FIG. 1. (Left) Illustrating large right lower lobe cavity. Note fluid level. Sputum repeatedly positive for tubercle bacilli.

FIG. 2. (Right) Lateral film of same patient. Illustrated are the posterior position of the lesion, the fluid level and the surrounding atelectasis of the dorsal segment in the lower lobe.

In addition to lateral views, oblique projections were especially valuable in demonstrating retrocardiac lesions. Here special use was made of a left anterior oblique film taken from 5 to 10 degrees from the postero-anterior axis. At this small angle the left cardiac border is superimposed on the left side of the spine while the retrocardiac lung field, including the entire left lower lobe, is thrown into clear vision. This view has certain advantages over the direct lateral or standard left oblique view because in these two positions a thin-walled cavity may be superimposed upon the spine and fail to be visualized for lack of sufficient density. The accompanying diagrams (figures 3, 4, and 5) illustrate this phenomenon. With this method, as with all special projections, the preliminary use of fluoroscopy is of great value. The lesions were distributed approximately equally between the right and left lungs and two were bilateral at the time of discovery.

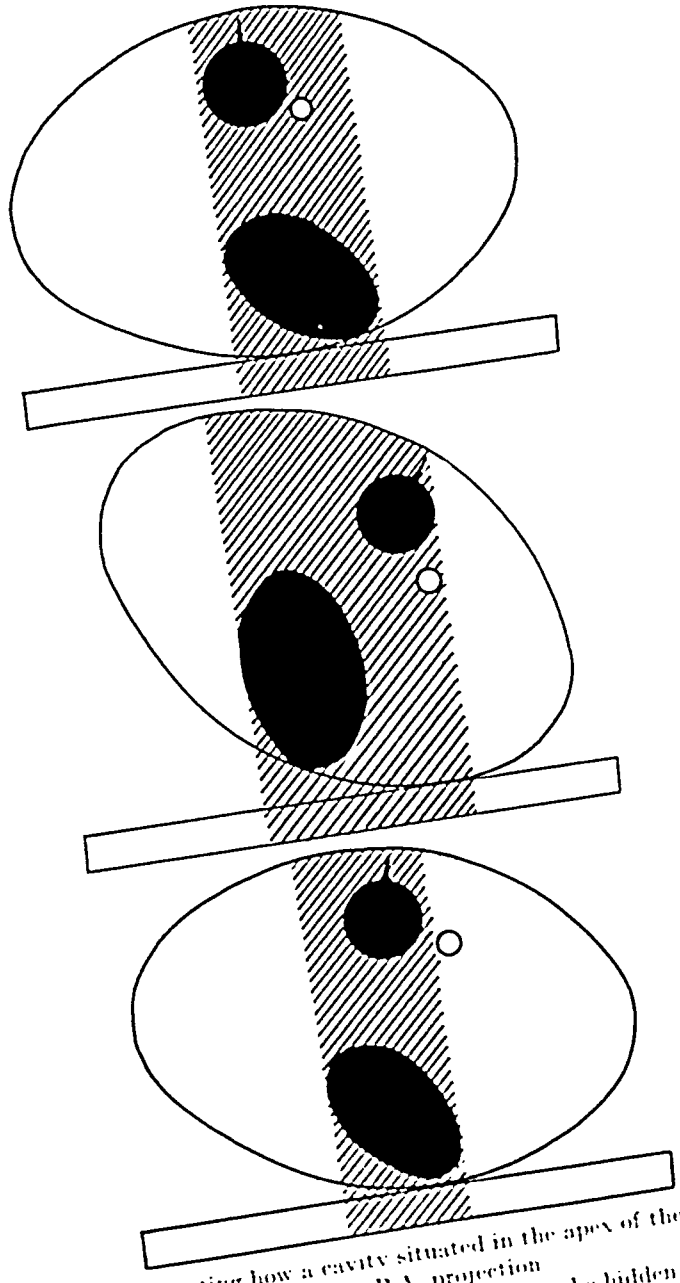

FIG. 3. (Top) Illustrating how a cavity situated in the apex of the left lower lobe may be hidden by the cardiac shadow on the P.A. projection

FIG. 4. (Center) Illustrating how the same cavity may be hidden by the shadow of the spine in the usual left oblique projection

FIG. 5. (Bottom) The cavity can be visualized by means of a left oblique film made about 15 degrees from the P.A.

Bronchial disease: An interesting observation, and one with diagnostic and therapeutic significance, was the high percentage of bronchial disease. The presence of any one of the following three criteria was accepted as diagnostic of sig-

nificant bronchial involvement: (*1*) bronchoscopic evidence of stenosis or severe tracheobronchitis; (*2*) roentgenographic evidence of atelectasis before collapse therapy or immediately after induction of pneumothorax; or (*3*) cavity behavior characteristic of a tension cavity.

Bronchial disease was believed to be present in 37 patients (75 per cent). Well over one-half of these had typical tension cavities, while 25 per cent each showed atelectasis or bronchoscopic findings. Many cases showed two or all three of these evidences of bronchial tuberculosis. The relatively small number of positive bronchoscopies was due in part to the fact that many patients were not bronchoscoped. Moreover, the distal bronchi only may have been involved and these would often be inaccessible to the bronchoscope. In a series of 50 patients admitted consecutively with disease in the upper lobes, the incidence of bronchial disease was considerably lower (7 cases) than in the lower lobe cases.

Clinical course: During the period of study many (60 per cent) of the lower lobe lesions showed evidences of extension. In most instances the extensions were to the upper lobes and were roentgenologically similar in appearance and in behavior to "spreads" found in primarily upper lobe cases. In a fair proportion the "spread" had occurred at the time of the first diagnosis, although earlier films in each case were available showing that the disease was primarily lower lobe at its start. In some, the "spreads" occurred within a few weeks of diagnosis while in others many years elapsed during which time the lesion remained confined to the lower lobe.

Prognosis: The average period of observation of the group was three years. It appears that the outlook for these lesions even to the point of arrest is only fair. Long term (2 to 5 years) follow-ups are not available in any of these inactive cases. Nevertheless, of the total number, 5 are dead, 19 are arrested, and the remainder are active. Some of the last named group are on the road to arrest while others will undoubtedly fail to recover. In this study the diagnosis of "arrested" was made by the most rigid criteria, *i.e.*, stable roentgenographic findings compatible with a healed lesion for well over the minimum of six months, numerous negative examinations for tubercle bacilli, including sputum and gastric cultures, and four to six months of walking exercise. It is understood and emphasized, however, that "arrest" in a previously cavitary lesion is not synonymous with "cure." Many upper lobe cavitary lesions "arrest" with subsequent relapse. Such instances are usually from a group who were treated with either no collapse measures or collapse therapy of only short duration. Since these lower lobe cases, by and large, became arrested by the aid of similar short term collapse measures, a very cautious attitude should be maintained as to the ultimate fate of these patients.

Treatment

No one form of treatment has proved eminently satisfactory and this study did not reveal the ideal treatment for lower lobe tuberculosis. These patients have had almost the entire spectrum of therapeutic measures that are applied by the phthisiologist.

Pneumothorax: Sixteen patients had artificial pneumothorax on the side of the lower lobe lesion. The results were bad in every case. They will be detailed in another report (4) but were characterized by atelectases, pleural effusions, unexpandable lungs, and persistently open cavities. In not a single case could pneumothorax be carried as a "clean" satisfactory collapse for two to three years with satisfactory re-expansion of the lung and eventual arrest of the disease.

Resection: Three patients were subjected to resection procedures. In 2 the diagnosis of neoplasm could not be excluded. In one patient the disease became arrested after lobectomy and in the other a similar result was observed after pneumonectomy. In the latter patient, however, extrapulmonary lesions appeared which are gradually responding to streptomycin and orthopedic surgery. The third resection, a lobectomy, was done in a patient who had attained an unsatisfactory result despite various combinations of phrenic surgery, pneumoperitoneum, pneumothorax, and streptomycin. A postoperative spread of disease to the other lung occurred and the ultimate prognosis does not seem favorable.

Thoracoplasty: Thoracoplasty was not used as a treatment for any lesion limited to a lower lobe. The operation was performed in 4 patients for the following indications: unexpandable lung (postpneumothorax), extensive ipsolateral upper lobe extension, contralateral upper lobe extension, and following pneumonectomy. In 3 of these patients the disease is arrested and in the fourth it is still active.

Cavernostomy: In 2 patients open cavernostomies have been performed through the posterior chest wall. In one case streptomycin plus surgery led to temporary cavity closure, and in the other cavernostomy was combined with one month of streptomycin followed by phrenic surgery and pneumoperitoneum. It is too soon to evaluate the treatment.

Streptomycin: Six cases received bed-rest plus streptomycin, unaccompanied by surgery. Two were treated for extensive "spreads" and died during treatment. Three, with tension cavities and "spreads", showed slight but insignificant improvement. In one patient the disease became arrested.

Phrenic nerve crush: Phrenic surgery was performed alone in 4 cases. It was combined with pneumoperitoneum in 13 cases and with pneumothorax in 5. Of those with phrenic crush alone, the disease remained active in one and became arrested in 3. With the combination of phrenic crush and pneumoperitoneum the disease became arrested in 7 and is still active in 6 patients. Phrenic surgery plus pneumothorax resulted in three arrested lesions while two remain active. In each case where phrenic surgery with or without pneumoperitoneum was effective, cavity closure occurred early (three to six months) with reversal of infectiousness and clearing of exudative lesions. Pneumoperitoneum in this group was continued for an average of two and one-fourth years. The optimum duration of this treatment is not known and only observation of this type of case for five or more years will reveal whether this period of pneumoperitoneum (two and a quarter years) is long enough to promote permanent healing. At the present time there must be considerable doubt about the permanence of healing of a large cavity with surrounding disease, in the absence of permanent collapse of the lung

area involved, despite early good results. Figures 6 and 7 illustrate a spectacularly successful result with phrenic surgery and pneumoperitoneum in the patient whose initial films are shown in figures 1 and 2.

Rest treatment: Twelve patients received bed-rest only. In 7 the lesions were still confined to the lower lobes when this treatment was started. Of these there were 5 good results (the basal lesion cleared or became inactive, although one required contralateral thoracoplasty for an extension of the disease) while 2 have remaining active lower lobe disease. Spontaneous cavity closure occurred in 3 of these 5 patients with good results, and clearing of the infiltrate occurred in the

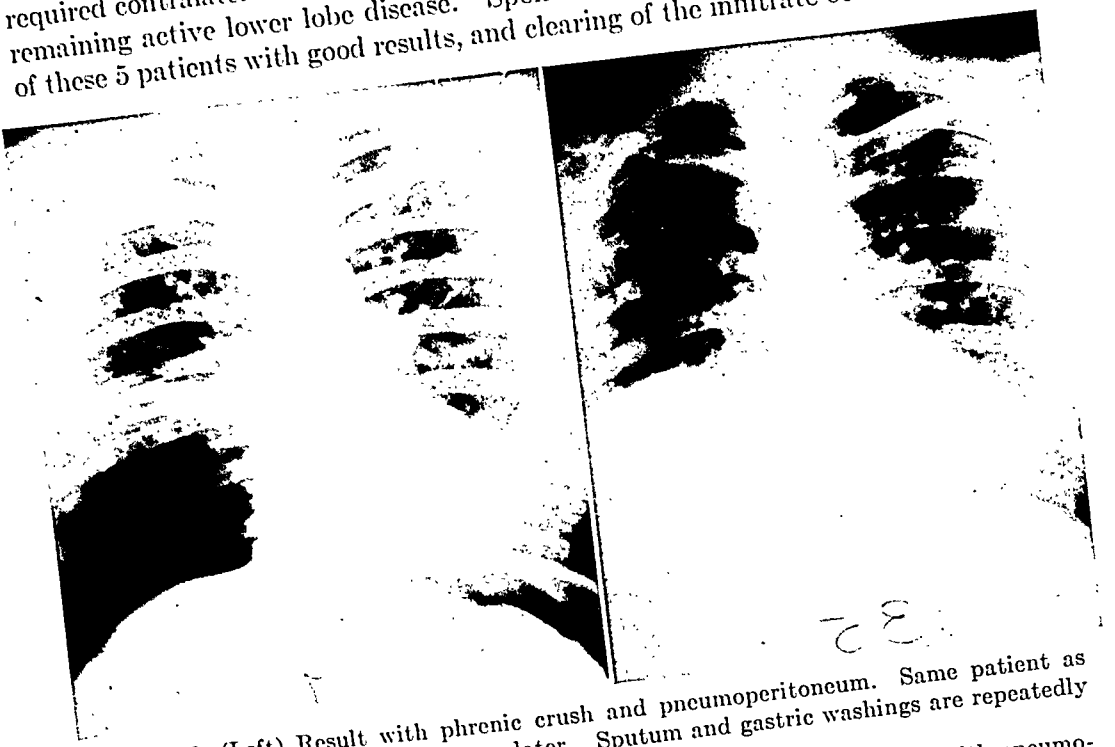

FIG. 6. (Left) Result with phrenic crush and pneumoperitoneum. Same patient as shown in figures 1 and 2, three years later. Sputum and gastric washings are repeatedly negative for tubercle bacilli on culture.

FIG. 7. (Right) Same case shown in figures 1, 2 and 6. Disease arrested with pneumoperitoneum abandoned after two and one-half years.

others. Such behavior is not uncommon with tension cavities, due perhaps to complete occlusion of the draining bronchus.

In summarizing the results of treatment these patients must be divided into two groups. One consists of those individuals who originally had lower lobe disease but in whom the disease has spread to such a point that treatment directed towards lower lobe tuberculosis could no longer be evaluated. The remainder (35 patients) had lesions confined to the lower lobe while undergoing treatment. By "good" results here are meant: (a) arrested tuberculosis, or (b) an inactive lesion on the lower lobe side despite continuing active contralateral upper lobe disease. The latter category was included as it is felt that lower lobe tuberculosis presents a mechanical as well as an immunological problem in cavity closure.

Rest alone was used in 7 cases, with good results in 5. Rest plus streptomycin gave good results in one case with poor results in 3 others. Phrenic crush alone yielded good results in 3 and poor in none. The results observed with phrenic crush plus pneumoperitoneum were good in 8 instances and poor in 3. Pneumothorax gave poor results in 13 patients and questionable results in 3 others. Pulmonary resection yielded good results in 2 cases and was of doubtful value in a third, and cavernostomy was followed by one good and one fair result. It may be seen from these figures that there is no "ideal" treatment plan revealed by study of this series but the observations point to certain tentative conclusions: (1) pneumothorax is contraindicated in this type of lesion; (2) rest, probably supplemented at an early date by phrenic nerve crush and pneumoperitoneum, offers the best chance of reaching arrest with relatively conservative measures; and (3) cavernostomy (with or without phrenic crush and pneumoperitoneum) and resections, supplemented by streptomycin, should be held in reserve until the above measures are unsuccessful.

Possible Relationship of Location of Lesion to Resistance to Tuberculosis

One question toward which this study was directed bears on the pathogenesis of lower lobe tuberculosis, in view of Dock's theory of upper lobe localization of this disease. In a carefully calculated mathematical demonstration, Dock has utilized newer knowledge of cardiovascular dynamics to prove that the circulation in the pulmonary apices is virtually nil in the erect position. The systolic pressure in the pulmonary artery is 20 to 25 mm. Hg, the diastolic pressure is 5 to 7 mm. and the average mean pulmonary arterial pressure as it leaves the heart is 15 to 18 mm. Hg. A column of blood 20 to 25 cm. in height exerts an equal (and opposite) pressure by its hydrostatic effect. The apex of the lung is 15 to 30 cm. cephalad to the pulmonary artery and therefore the pulmonary arterial pressure in the apex of the lung, with the patient seated or standing, is rarely over 10 mm. and in tall long-chested individuals it should be zero. The osmotic tension of the blood proteins is 25 mm. Hg and that of the tissue proteins is 10. There exists, therefore, an osmotic force of 15 mm. Hg which tends to keep fluids within the blood vessels and the systolic pressure must exceed this to produce an effective intra-arterial pressure which will force fluids (with oxygen, nutrients, antibodies, etcetera) into the tissues. Because of these facts Dock draws the following conclusions: (1) Little or no blood flows through the upper one-third of the lungs during two-thirds of the day. (2) No tissue fluid or lymph is formed there while the patient is in the erect position. (3) Little or no carbon dioxide or oxygen exchange can take place nor can antibodies be formed nor can removal of bacteria or dilution of their products take place. Dock suggests that tubercle bacilli are inhaled into all parts of the lungs but host resistance is usually great enough to destroy them in those sections of the lungs having adequate blood supply. In the apices of the upper lobes, however, for the reasons given above, the forces of host resistance are reduced in efficiency.

Why then does lower lobe tuberculosis develop? Dock reasons that, as most people can destroy tubercle bacilli which find their way to the lower lobes, the

development of active disease in these areas must be a manifestation of an inherently lowered resistance. Such patients, he feels, should not be benefited by bed-rest, which, he believes, may exert its desirable effect in upper lobe tuberculosis largely by virtue of the increased blood pressure produced in the involved area when the patient is recumbent.

Each one of the present series of 48 patients was studied from the standpoint of whether an unusually low resistance to tuberculosis was evident. It must be remembered that in no case was the lesion located above the level of the main pulmonary arterial shadow on the erect postero-anterior roentgenogram. It is understood that no predictable course can be expected in the individual case of tuberculosis but it is believed that in a group of this size an approach can be made to the problem. The following criteria were set up for evidence of lowered resistance: (*a*) steady spreading disease with little fibrosis and healing; and (*b*) rapid clinical downhill course with febrile episodes and progressive caseation.

Of the 48 cases analysis reveals that only 5 showed evidence of decreased resistance when these criteria were applied to each one individually.

The subsequent course of the group is in agreement with this evaluation. Of 48 patients (of whom 46 had moderately or far advanced disease at the time of admission to this hospital) followed for an average of three years, only five (10 per cent) are dead and 40 per cent have arrested lesions. Most of the patients with arrested lesions reached this clinical status without major collapse measures. Bed-rest, phrenic crush and pneumoperitoneum accounted for all but 4 of the arrested lesions.

The results observed in this group of patients do not appear to support the validity of the idea that patients with lower lobe disease have inherently lowered resistance to tuberculosis and hence develop the disease in this atypical location.

In the other series of cases studied (2, 3) the same conclusion was reached, *i.e.*, that the clinical course and prognosis in lower lobe disease parallels that of tuberculosis in the more usual distribution. It seems probable that poor results with therapy, which give rise to the impression that disease in this location is refractory to treatment, are due to mechanical factors rather than immunological ones. The high percentage of bronchial disease found in this series plus the related bad results with pneumothorax play a large part. In addition there exists the reluctance of the surgeon to perform thoracoplasty in cases where the upper one-half of the lung field is normal. There seems no evidence to substantiate Dock's theory that this localization is in itself evidence of lowered resistance or immunity. This is not, of course, an effort to support or disprove Dock's basic analysis of the vasodynamics of the lung and its relationship to upper lobe localization.

SUMMARY

1. Lower lobe tuberculosis is a fairly frequent problem in the diagnosis and treatment of pulmonary disease.

2. It is often misdiagnosed when first seen clinically and radiologically. Some of the reasons for this are discussed. Repeated sputum studies for tubercle bacilli, including cultures and gastric lavage, as well as repeated roentgenograms

should be obtained in the presence of lower lobe inflammatory lesions which fail to clear rapidly and completely. Lateral and oblique roentgenograms may be of value in clarifying the diagnosis. The possibility of tuberculosis cannot be dismissed as long as roentgenographic findings persist.

3. Most patients with lower lobe tuberculosis have bronchoscopic or radiologic evidence of bronchial disease. This must be considered in the diagnostic and therapeutic management of these cases.

4. Artificial pneumothorax almost always gives unsatisfactory results in this type of lesion.

5. No ideal type of therapy is demonstrated by study of this series. It seems probable that a short period of bed-rest followed by phrenic crush and pneumoperitoneum will offer the best results. Even this, however, is frequently unsuccessful. In addition, little is known of the ultimate fate of the patients who become arrested under this regimen.

6. The status of such measures as resection and cavernostomy plus streptomycin, either as primary treatment or in refractory cases, has as yet to be sufficiently explored.

7. Patients with lower lobe tuberculosis apparently do not have decreased resistance to this disease. The prognosis is approximately the same as that of patients with disease of similar extent in the upper lung fields. Difficulties in treatment would appear to be mechanical rather than immunological.

SUMARIO

Tuberculosis Pulmonar del Lóbulo Inferior

1. La tuberculosis del lóbulo inferior del pulmón constituye un problema bastante frecuente en el diagnóstico y tratamiento de las neumopatías.

2. Al ser observada por primera vez clínica y radiológicamente, es a menudo diagnosticada erróneamente, y aquí se discuten algunas de las causas de ello. En presencia de lesiones inflamatorias del lóbulo inferior que no desaparecen rápida y completamente, hay que hacer repetidos estudios del esputo, incluso cultivos y lavado gástrico, en busca de bacilos tuberculosos, así como radiografías repetidas. En el esclarecimiento del diagnóstico las roentgenografías laterales y oblicuas pueden resultar útiles. En tanto que persistan los hallazgos radiográficos no cabe descartar la posibilidad de tuberculosis.

3. La mayor parte de los enfermos con tuberculosis del lóbulo inferior muestran signos broncoscópicos o radiológicos de afección bronquial, lo cual hay que considerar en la atención diagnóstica y terapéutica de estos casos.

4. En esta forma de lesión el neumotórax terapéutico casi siempre da resultados poco satisfactorios.

5. El estudio de esta serie no ha revelado ninguna terapéutica ideal. Parece probable que un breve período de descanso en cama seguido de la trituración del frénico y el neumoperitoneo es lo que ofrece más promesa, y aun esto frecuentemente no da resultado. Además, poco se sabe del destino definitivo de los enfermos que se estacionan con este régimen.

6. Está aun por explorar a fondo el estado de medidas tales como la resección

y la cavernostomía, unidas a la estreptomicina, ya como tratamiento primario o en los casos refractarios.

7. Los enfermos con tuberculosis del lóbulo inferior aparentemente no muestran menor resistencia a la enfermedad. El pronóstico es aproximadamente idéntico que en los que tienen enfermedad de proporciones semejantes en los campos superiores del pulmón. Las dificultades terapéuticas parecen ser mecánicas más bien que inmunológicas.

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POOR RESULTS WITH ARTIFICIAL PNEUMOTHORAX IN LOWER LOBE TUBERCULOSIS[1, 2]

EMIL ROTHSTEIN

In a recent study of lower lobe tuberculosis (1), two related observations were made: The incidence of bronchial disease was unusually high and the results with artificial pneumothorax were very poor. In this paper these results are considered in detail.

CLINICAL MATERIAL

Seventeen patients were studied. One, however, had bilateral pneumothorax for bilateral lower lobe cavitation so an analysis of 18 pneumothoraces will be made. All of the patients were males. The average age was 28 years. Four were Negroes and the remainder were white. A positive bacteriologic diagnosis of tuberculosis was made in each case. Ten of the lesions were in the right lower lobe and eight in the left lower lobe. The average time between the first appearance of the lesion on the roentgenograms and the induction of the pneumothorax was eight months.

Description of the lesions: The lesion was moderately advanced in each instance at the time of induction of the pneumothorax. In all but 2 cases definite cavitation was visible and in the 2 exceptions the lesion was pneumonic in character with cavitation presumed but not demonstrated radiologically. In the cases showing excavation, the cavity was in the apex of the lower lobe (appearing to be hilar on the postero-anterior projection) in 7 and lower in the lung field in 9. The upper lung fields were essentially normal in all cases or presented a recent minimal extension of the lower lobe tuberculosis which was of little immediate significance.

Bronchial involvement: Direct or indirect evidences of bronchial involvement were present in all but one case. The following findings were considered to indicate tuberculous disease of the draining bronchus or bronchi: (*1*) bronchoscopic findings of stenosis or inflammation in the involved lobe; (*2*) partial atelectasis of the involved lobe either prior to or immediately after the induction of the pneumothorax; or (*3*) cavity changes consisting of fluid levels or fairly rapid changes in the size and the shape of cavities. Most of these last mentioned findings occurred in typical tension cavities which were round or oval with clear inner borders and relatively normal peri-cavitary lung fields. Twelve of the cavities presented fluid levels while sudden changes in size were recorded in 13. Atelectasis of a segment of the lower lobe was present in 5 cases before pneumothorax and in 5 immediately after its induction. Bronchoscopic changes were noted in 6 of the 9 cases in which bronchoscopy was performed. Thus it can be seen that many patients presented two or three of these manifestations.

[1] From the U. S. Veterans Administration Hospital, Tuberculosis Division, Wood' Wisconsin.

[2] Published with permission of the Chief Medical Director, Department of Medicine and Surgery, Veterans Administration, who assumes no responsibility for the opinions expressed or conclusions drawn by the author.

RESULTS WITH ARTIFICIAL PNEUMOTHORAX

The goal to be achieved with artificial pneumothorax is arrest of the tuberculous process, leaving a lung which re-expands and has a fairly good residual respiratory function. Serious pleural complications must be absent. The writer believes, with most other phthisiotherapists, that pneumothorax should be a reversible procedure which conserves much of the function of the involved lung. It is thought that a fibrothorax with a nonfunctioning lung, or an unexpandable lung with permanent pneumothorax or persistent effusion, must be considered a poor result even if the disease in the underlying lung is apparently inactive. It is on the basis of this evaluation that the following analysis was made.

In none of the 18 cases was a satisfactory result obtained (table 1). In 6 cases inoperable adhesions led to the discontinuation of the pneumothorax after an average period of six months. In 4 cases pneumothorax was stopped after four

TABLE 1

Reasons for poor results in pneumothorax cases of lower lobe tuberculosis

Inoperable adhesions	6
Massive recurrent effusions	5
Persistent cavity without adhesions	4
Voluntary discontinuation	1
Unexpandable lung	1
Progressive atelectasis	1
Total	18

to six weeks; 2 pneumothoraces were continued in other hospitals for twelve and twenty-one months, respectively. One patient has had lobectomy and 2 are scheduled for thoracoplasty but all 6 with inoperable adhesions still have active tuberculosis.

In 5 cases persistent massive pleural effusions led to the cessation of pneumothorax after an average period of eleven months of collapse. Fluid appeared on an average of five months after the onset of collapse therapy. In 2 of these patients acid-fast bacilli were present in the fluid but no purulent empyema developed. One patient has had thoracoplasty for an unexpanded lung, a second is undergoing thoracoplasty for an ipsolateral upper lobe cavity which developed subsequently, a third still has active disease, and the other two have arrested lesions. Both of the latter have badly crippled shrunken lungs due to inadequate expansion and fibrothorax. Unfortunately, the last statement is supported only by clinical observations as bronchospirometric readings are not available in either case.

In 4 cases open cavitation persisted despite the absence of evident adhesions. Pneumothorax was discontinued after an average period of fourteen months in this group. In general, the treatment was continued for such a long period because of cavities which would be lost to view temporarily, only to reappear six or eight months later. One of these lesions became arrested following phrenic crush and pneumoperitoneum. In a second case cavity closure apparently occurred following open cavernostomy and streptomycin. A third patient had thoraco-

plasty for further ipsolateral extension while the fourth still has active disease. In one of these cases a tension cavity increased considerably in size while pneumothorax was being administered.

Three miscellaneous causes of failure were as follows. In one case unexpandable lung accompanied by persistent effusion necessitated thoracoplasty. This patient's disease then became arrested. In a second case progressive atelectasis developed along with a pleural effusion which contained tubercle bacilli. The pneumothorax was discontinued and a phrenic crush was performed with subsequent arrest of the disease. The third patient voluntarily discontinued the treatment three months after apparent cavity closure. The cavity reopened one year later and the disease spread to both lungs.

The present status of the series is as follows: In 5 patients the disease is arrested, in 2 with pneumothorax alone. In both of these the result is considered unsatisfactory because of shrunken lung plus fibrothorax. The disease has become arrested in an additional 3 patients after thoracoplasty, phrenic crush, and phrenic crush plus pneumoperitoneum, respectively. The remaining 13 patients in the series all have persistent active tuberculosis.

SUMMARY

Eighteen lower lobe tuberculous lesions were treated with artificial pneumothorax. All were in males and the lesions were all moderately advanced. All but 2 were frankly cavitary. Only 2 became arrested with artificial pneumothorax therapy alone and in these instances the patients were left with shrunken lungs and thickened pleura. Three of the remaining lesions became arrested with other methods of treatment and the 10 others are still active.

It is believed that the poor results observed are in part due to the high incidence of bronchial disease as shown by bronchoscopy or by secondary pulmonary changes.

SUMARIO

Malos Resultados del Neumotórax Terapéutico en la Tuberculosis del Lóbulo Inferior

Dieciocho lesiones tuberculosas del lóbulo inferior fueron tratadas con el neumotórax artificial. Todas eran en varones y todas eran moderadamente avanzadas. Todas, menos dos, eran netamente cavitarias. Sólo dos, o sea 11.1 por ciento, se estacionaron bajo la acción exclusiva del neumotórax terapéutico, y en esos casos los enfermos quedaron con pulmones contraídos y pleuras espesadas. Tres de las lesiones restantes se estacionaron con otras técnicas terapéuticas, y las otras se hallan todavía activas.

Parece que los malos resultados observados se deben en parte a la alta incidencia de afección bronquial (95 por ciento) según revelaran la broncoscopia o las alteraciones pulmonares secundarias.

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PROGNOSIS OF INSPISSATED CAVITIES[1,2]

ROBERT S. STUDY[3] AND PHILIP MORGENSTERN[4]

INTRODUCTION

What is the prognosis of the inspissated cavity in tuberculosis? Several writers during the past ten years (9, 10, 12, 16, 17) have presented pathological and physiological evidence that inspissation is not an uncommon method of cavity healing. In spite of this fact, the idea has persisted among most chest physicians that an inspissated cavity is a dangerous, unstable lesion which may discharge its contents into the bronchus at any time. In the English literature only Shamaskin (14) has ventured the opinion that an inspissated cavity is a retrogressing or healed lesion and should be treated conservatively. He based his conclusions on a study of six cases.

Since the appearance of Shamaskin's article in 1941 (14), the question of prognosis in an inspissated cavity with negative sputum has assumed increased importance. This is due largely to advances in thoracic surgery, particularly in pulmonary resection. Many thoracic surgeons now regard the inspissated cavity as a definite indication for lobectomy. Is this attitude justified? The present study was undertaken to clarify the prognosis of the inspissated cavity and to evaluate the indication for lobectomy in such cases.

CASE REPORTS

A survey of approximately 1,000 patients now in the hospital or recently discharged and being followed in the outpatient department revealed 24 cases in which inspissated cavities were present. Under this heading were included only those cases where serial roentgenograms revealed a definite open cavity which became filled with secretions, was transformed into a dense opacity, and then began to decrease in size as the process progressed. All but 2 of the cases showed acid-fast bacilli in the sputum at the time they came under our observation.

In table 1 may be seen the pertinent data concerning the course of inspissation of the cavities in 24 patients who have been observed for periods ranging from six months to seven and one-half years. In the following paragraphs the clinical course of the first 6 patients shown in the table are described in detail. Serial chest roentgenograms illustrate the typical changes occurring in cavities during the process of "inspissation."

Case 1 (A. D. H.): The onset of this patient's tuberculosis was in September 1942. The admission film at Oteen on November 19, 1942 (figure 1) revealed disease in the basal

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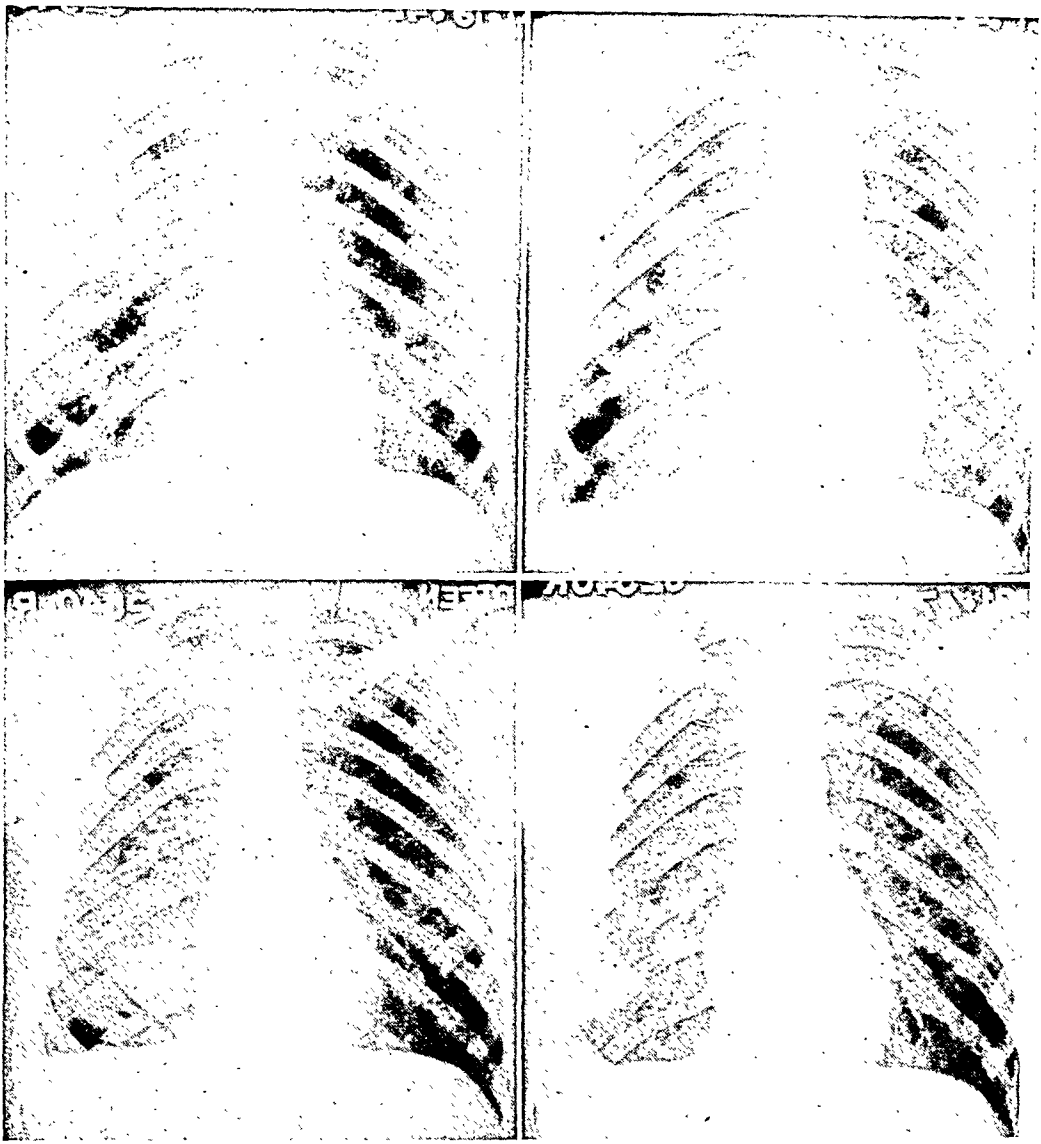
TABLE 1

NUMBER	NAME	ONSET	COLLAPSE THERAPY	CAVITY FIRST SEEN	CAVITY "INSPISSATED"	BACTERIOLOGY		
						Date of last sputum positive for tubercle bacilli	Subsequent sputum examination	
							Negative microscopics	Negative cultures
1.	A. D. H.	Sept. 1942	Pnx.† 12/ 2/42 8/14/46	11-19-42	1- 1-43	1-18-43	44	0
2.	H. I.	Dec. 1927	Phr.* 1932 Pnx. 1932-33	1-20-40	11-28-40	5-28-40	38	3
3.	J. T. U.	Mar. 1943	None	10- 7-43	1-26-46	9-27-44	26	4
4.	W. E. D.	Feb. 1943	Pnx. 11-19-43 3-25-47 Phr. 4- 4-47	9-18-43	7-21-44	2- 5-44	32	4
5.	T. A.	1941	Phr. 6-22-46 Pnm.‡ 8-13-46	4-12-44	8- 8-46	5-27-45	11	7
6.	W. W.	June 1946	Phr. 9-11-46	7-19-46	12-12-46	None	13	5
7.	T. F.	May 1942	Phr. 5-5-44, 2-8-45, 5-15-45 Pnx. 10-2-45	9- 6-43	12-14-45	5-17-45	10	8
8.	C. L.	Apr. 1944	Pnx. 7-8-44 2-5-46	4-11-44	3-14-45	7-12-44	22	6
9.	J. E.	Mar. 1943	None	5-14-45	10-23-45	5-10-45	5	4
10.	L. S. A.	Apr. 1943	Phr. 6-2-44 and 7-4-45 Pnx. 2-8-45 6-20-45 Pnm. 8-9-44 3-10-45 and 8-6-46 to date	6-28-44	1-10-47	3-20-46	5	4
11.	E. A. S.	Nov. 1945	Phr. 3-12-46 Pnm. 5-1-46 to date	2-11-46	lower cavity inspissated 5-24-46, upper on 8-14-46	5-24-46	6	3
12.	P. F.	Apr. 1946	Phr. 7-22-46 Pnm. 8-13-46; 9-20-46	6-22-46	9-20-46	7-12-46	10	3
13.	J. B.	July 1946	Phr. 9-2-46 and 7-22-47	8-12-46	3- 3-47	None Positive	6	1
14.	D. H.	Jan. 1946	Pnx. 9-24-46 to date Pneumonolysis 5-9-47	8- 7-46 2 cavities	Lower cavity inspissated first; upper inspissated 5-14-47	7-10-46	12	2

TABLE 1—*Continued*

NUMBER	NAME	ONSET	COLLAPSE THERAPY	CAVITY FIRST SEEN	CAVITY "INSPISSATED"	BACTERIOLOGY: Date of last sputum positive for tubercle bacilli	BACTERIOLOGY: Subsequent sputum examinations: Negative microscopics	BACTERIOLOGY: Subsequent sputum examinations: negative cultures
15.	J. A. S.	Mar. 1946	Pnx. 8-6-46 to date	6-17-46	8-16-46	8-20-46	14	6
16.	W. G.	Oct. 1946	Phr. 12-5-46 Pnm. 1-8-47 to date	11-21-46	2- 7-47	11-26-46	12	7
17.	R. H.	Nov. 1945	Pnx. 1-8-47 1-31-47 Pnm. 2-4-47 5-26-47 Phr. 2-14-47 Pregnancy delivered 9-9-47	12-27-46	2- 6-47	1-10-47	16	2
18.	C. P.	May 1943	Pnx. 2-21-44 4-4-44 Phr. 10-26-45 Streptomycin for endobronchial tuberculosis	1-29-44	1-30-46	2-18-47	6	3
19.	A. G. E.	Aug. 1946	Phr. 11-26-46 Pnm. 12-4-46 to date	10-23-46	1-14-47	3-22-47	11	5
20.	J. A. G.	Oct. 1946	Phr. 2-11-47 5-5-47 Pnm. 12-10-46	11-19-46 Two cavities	Upper cavity inspissated 4-2-47 and lower inspissated 7-16-47	4-7-47	4	0
21.	G. B.	Oct. 1946	None	3-14-47	7- 9-47	5-14-47	21	1
22.	A. S.	Nov. 1946	Phr. 5-13-47 Pnm. 6-30-47 to date	12-23-46	6-23-47	5- 1-47	9	0
23.	R. C.	Jan. 1947	Pnx. 6-12-47 to discharge	5-20-47	6-23-47	5-27-47	1	0
24.	G. D.	Feb. 1947	Pneumonolysis 8-6-47 Pnx. 5-6-47 to date Pneumonolysis 7-8-47	4- 2-47	6-10-47	4-23-47	10	0

* Phr. = phreniclasia.
† Pnx. = Pneumothorax.
‡ Pnm. = pneumoperitoneum.



FIGS. 1–4

FIG. 1. (upper left) Case 1. (A. D. H.). Admission film of November 19, 1942, showing disease in basal portion of right upper lobe with a 2 cm. cavity in second anterior interspace.

FIG. 2. (upper right) Case 1. Chest film, January 1, 1943, one month after institution of pneumothorax. The cavity is seen replaced by an irregular nodular density.

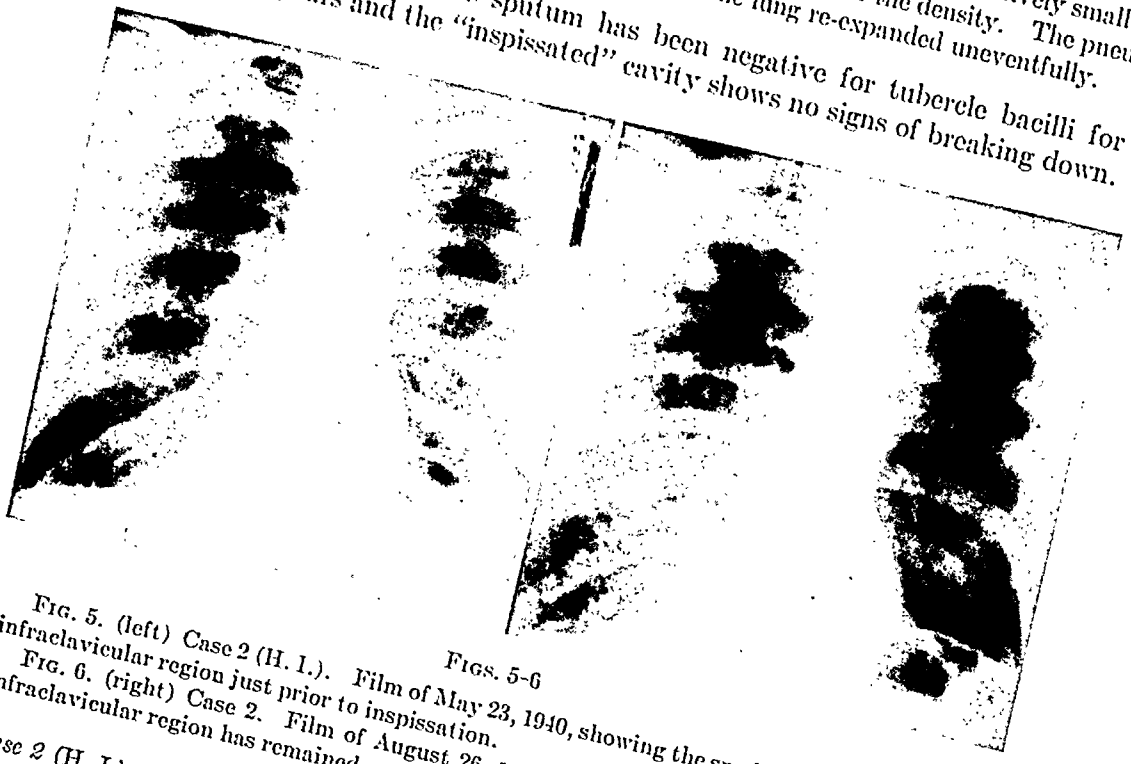
FIG. 3. (lower left) Case 1. Film of August 14, 1946. The nodular density representing a presumably inspissated cavity has become considerably smaller since 1943.

FIG. 4. (lower right) Case 1. Film of July 21, 1947. The lung is now completely re-expanded and the nodular density in the right infraclavicular region shows beginning calcium deposition.

portion of the right upper lobe with a 2 cm. cavity in the second anterior interspace. The sputum was positive for acid-fast bacilli. A right pneumothorax was instituted

December 2, 1942, and the sputum became negative for tubercle bacilli in February 1943. The film on January 1, 1943 (figure 2) showed that the cavity had been replaced by a nodular density interpreted as an inspissated cavity. On serial films through 1947 (figures 3, 4) it was noted that the "inspissated" cavity had become progressively smaller with deposition of some calcium deposits in the outer portions of the density. The pneumothorax was discontinued August 14, 1946, and the lung re-expanded uneventfully.

Comment: This patient's sputum has been negative for tubercle bacilli for almost five years and the "inspissated" cavity shows no signs of breaking down.



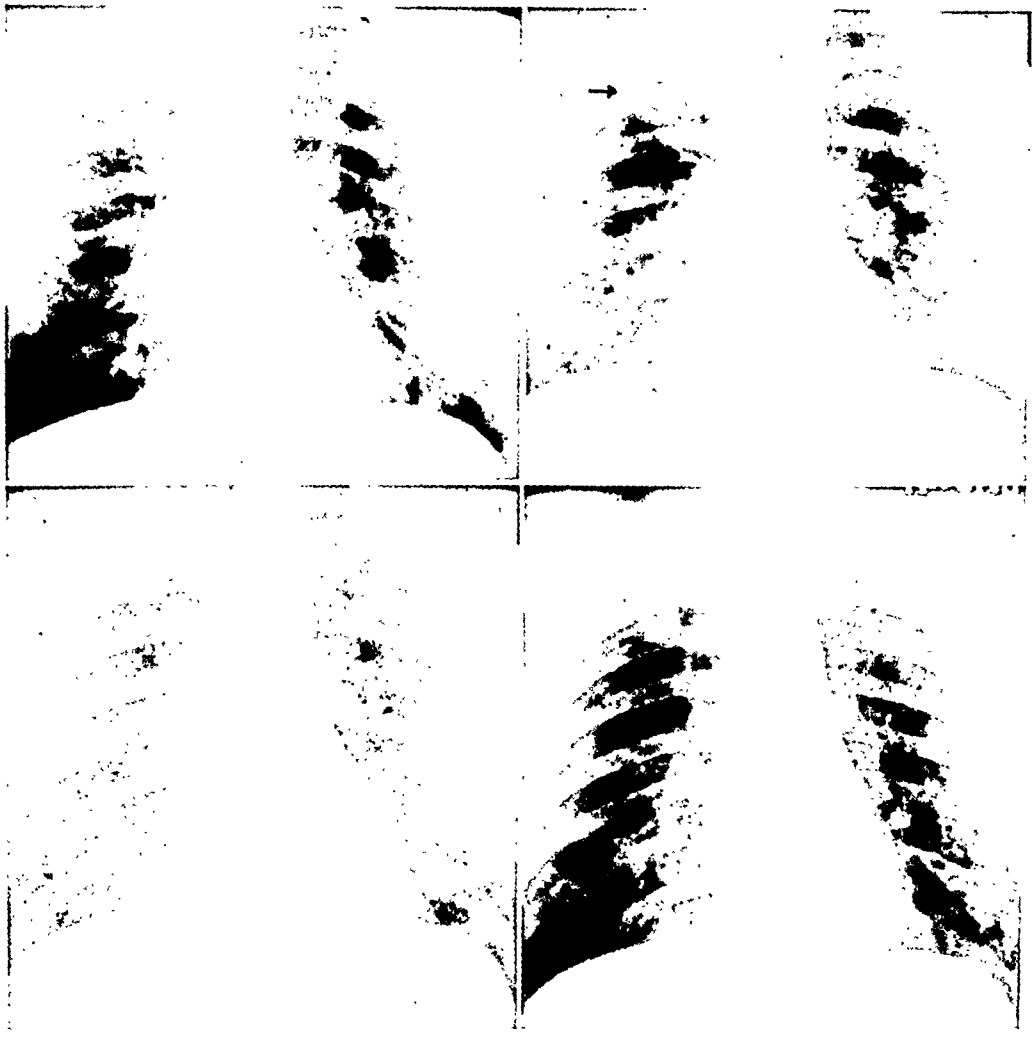
FIGS. 5-6

FIG. 5. (left) Case 2 (H. I.). Film of May 23, 1940, showing the small cavity in the right infraclavicular region just prior to inspissation.

FIG. 6. (right) Case 2. Film of August 26, 1947. The inspissated cavity in the right infraclavicular region has remained unchanged over a period of seven years.

Case 2 (H. I.): This patient was found to have tuberculosis in 1927. During the next thirteen years he was admitted to several tuberculosis sanstoriums. He was admitted to Oteen, January 19, 1940, with the diagnosis of moderately advanced pulmonary tuberculosis. His sputum was positive for acid-fast bacilli and a small thick-walled cavity was seen behind the second anterior rib on the right on January 20, 1940. Previous therapy consisted of a left phreniclasia in 1932 and a left pneumothorax in 1932–1933. His last sputum positive for tubercle bacilli was on May 28, 1940. The cavity was last seen May 23, 1940 (figure 5) and it was interpreted as "inspissated" on November 28, 1940. He was discharged as an arrested case in September 1943. Serial roentgenograms from November 1940 through August 26, 1947 (figure 6) have shown no appreciable change in the appearance of the "inspissated" cavity. He has had two admissions to Oteen since 1943, one for chronic bronchitis, the other for an upper respiratory infection. All sputum studies have remained negative for tubercle bacilli.

Comment: This case of an "inspissated" cavity has been followed for seven years and shows no tendency to exacerbation of the disease.



FIGS. 7–10

FIG. 7. (upper left) Case 3 (J. T. U.). Film of July 11, 1944, showing the large cavity in the right upper lobe with a basal fluid level.

FIG. 8. (upper right) Case 3. Film of December 6, 1944. The cavity in the right upper lobe is smaller, and the fluid level (indicated by arrow) is higher.

FIG. 9. (lower left) Case 3. Film of January 26, 1946. The cavity in the right upper lobe has been completely replaced by a uniform oval density which presumably represents an inspissated cavity.

FIG. 10. (lower right) Case 3. Film of June 19, 1947. The inspissated cavity is smaller and more sharply delineated.

Case 3 (J. T.U.): The onset of this patient's tuberculosis was in March 1943. Admission film of October 7, 1943, at Oteen showed a thick-walled cavity 5 by 3.5 cm. in the right infraclavicular region. The sputum was positive for acid-fast bacilli. A roentgenogram of July 11, 1944 (figure 7) showed extension of the disease to the left lower lung field. A fluid level was seen within the cavity in the right upper lobe and by December 6, 1944 (figure 8) the cavity had become definitely smaller with a thicker wall and a higher fluid

level. The last sputum positive for tubercle bacilli was on September 27, 1944. A chest film of January 26, 1946 (figure 9) was believed to show complete "inspissation." Serial roentgenograms taken through 1946 and 1947, the last on June 19, 1947 (figure 10), showed progressive contraction of the "inspissated" cavity with the formation of a hard nodular focus. The patient was discharged from the hospital on June 25, 1947.

Comment: This is a case of an "inspissated" cavity followed for over three years. The resulting nodular density has become progressively smaller. His sputum became noninfectious sixteen months before complete "inspissation" was demonstrated by roentgenography. During a period of more than three years examination of this patient's sputum with 14 direct smears, 12 concentrates, three cultures and one guinea pig inoculation has revealed no tubercle bacilli.

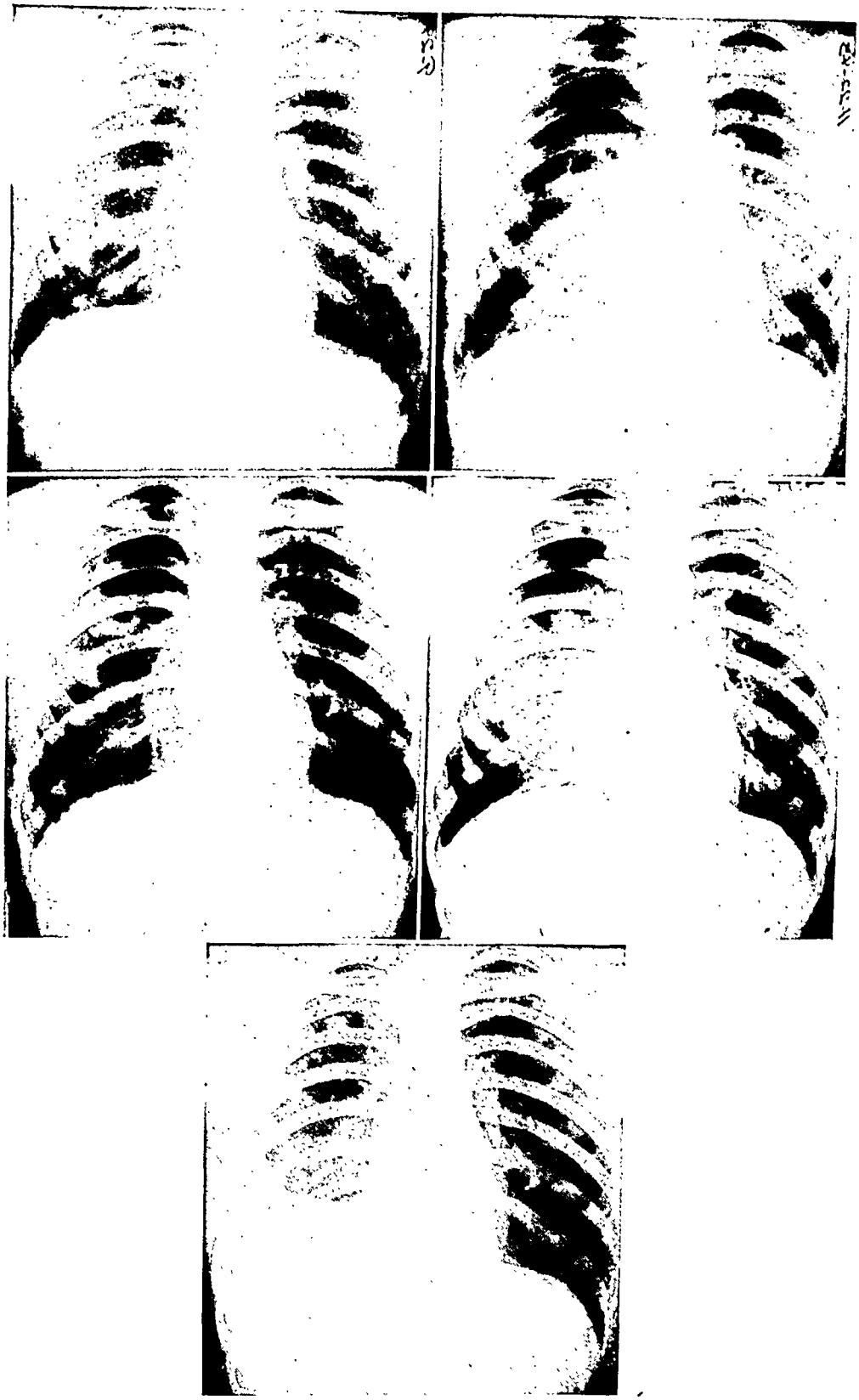
Case 4 (W. E. D.): The onset of this 24-year-old patient's tuberculosis was in February 1943. On September 29, 1943 (figure 11), a 3 cm. cavity with a basal fluid level was demonstrated in the right infraclavicular region by roentgenography. The sputum was positive for acid-fast bacilli and a right pneumothorax was instituted in November 1943 (figure 12). The patient was admitted to Oteen on January 3, 1944. By February 1, 1944, the cavity had diminished in size and was one-third filled with fluid (figure 13). His last sputum positive for tubercle bacilli was in February 1944, and by July 1, 1944, the cavity had been replaced by an oval density. In serial films taken on August 9, 1946 and August 22, 1947 (figures 14, 15), the stationary character of this nodular focus is demonstrated. This patient was discharged in September 1945, and the pneumothorax was discontinued in March 1947. Numerous sputum examinations since February 1944 have all been negative for tubercle bacilli.

Comment: This patient's sputum has been negative for tubercle bacilli for over three and one-half years and complete "inspissation" has been observed for approximately three years.

Case 5 (T. A.): The onset of this patient's tuberculosis was in 1941. He was admitted to Oteen April 11, 1944. On roentgenography a thick-walled cavity with a diameter of 4 by 6 cm. was seen in the left upper lobe and there was fibrocaseous involvement in the upper one-third of the right lung. The left upper lobe cavity remained unchanged in size (figure 16) until March 2, 1945, when it became smaller and showed a definite fluid level (figure 17). The patient's last sputum positive for acid-fast bacilli was May 27, 1945. Planigrams on June 22, 1946 (figure 18) showed the cavity to be still present. By August 8, 1946, the cavity was believed to be completely "inspissated." Films taken throughout the rest of 1946 and during 1947 showed the "inspissated" cavity to have become smaller, and a planigram on July 19, 1947 (figure 19) verified the stable character of the lesion. Since May 27, 1945 examination of the sputum (eight concentrates, seven cultures) has failed to reveal tubercle bacilli.

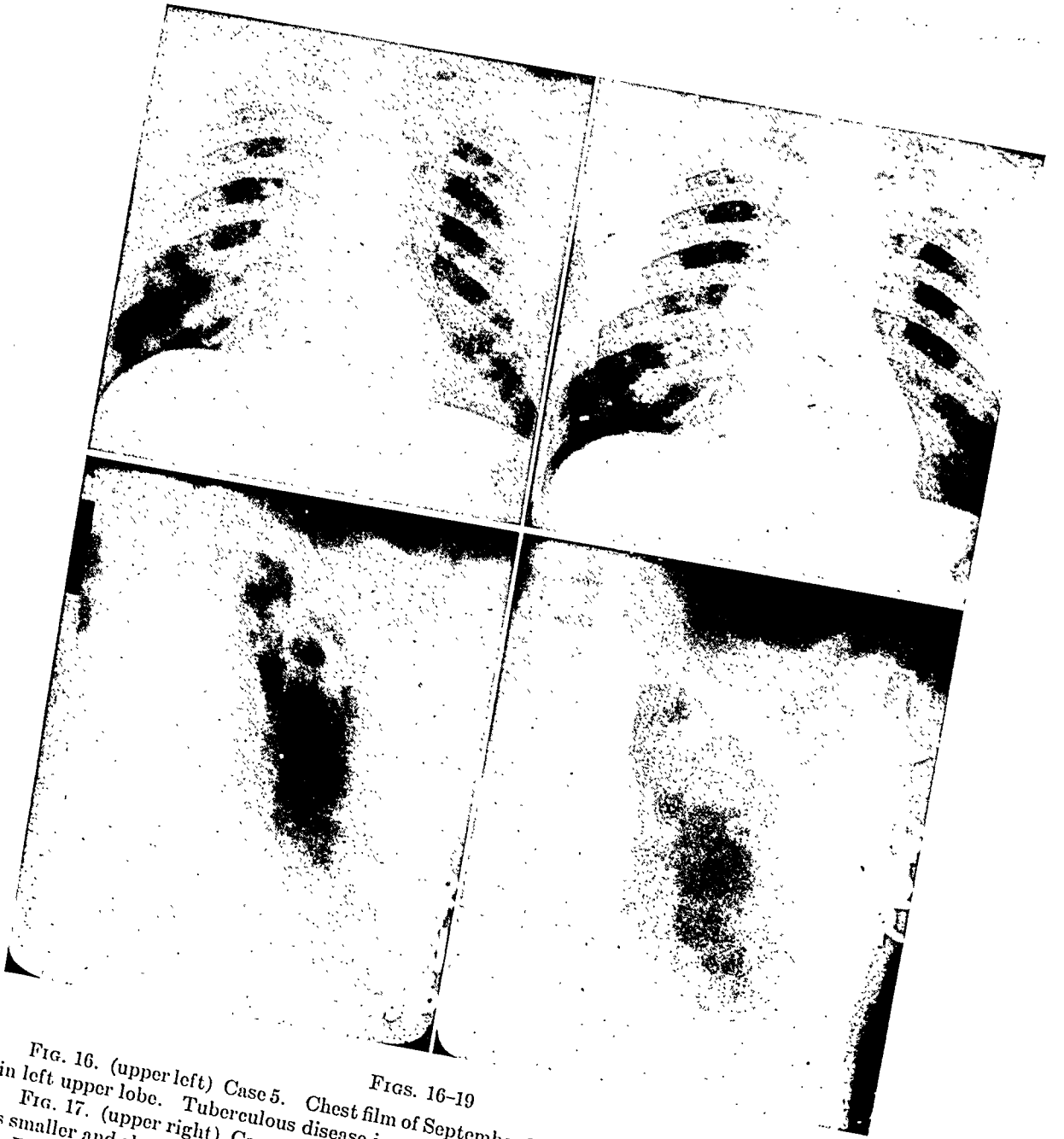
Comment: This patient's sputum has been negative for tubercle bacilli for two and one-half years and he is now on an ambulant status preparatory to being discharged.

Case 6 (W. W.): The onset of this patient's tuberculosis was in June 1946. At the time of his admission to Oteen on July 19, 1946, roentgenography revealed a small thick-walled cavity behind the left first anterior rib (figure 20). Numerous sputum studies failed to reveal acid-fast bacilli. The diagnosis of chronic pulmonary tuberculosis was based on



FIGS. 11–15

FIG. 11. (upper left) Case 4 (W. E. D.). Chest film of Sept. 29, 1943 shows a 3 cm. cavity with a basal fluid level in the right infraclavicular region.



FIGS. 16–19

FIG. 16. (upper left) Case 5. Chest film of September 28, 1944, showing 4 by 6 cm. cavity in left upper lobe. Tuberculous disease is present also in right upper lobe.

FIG. 17. (upper right) Case 5. Chest film of March 2, 1945. Cavity in left upper lobe is smaller and shows basal fluid level.

FIG. 18. (lower left) Case 5. Planigram of June 22, 1946 shows the contracted cavity in the left upper lobe. Sputum had already been negative for tubercle bacilli for one year at this time.

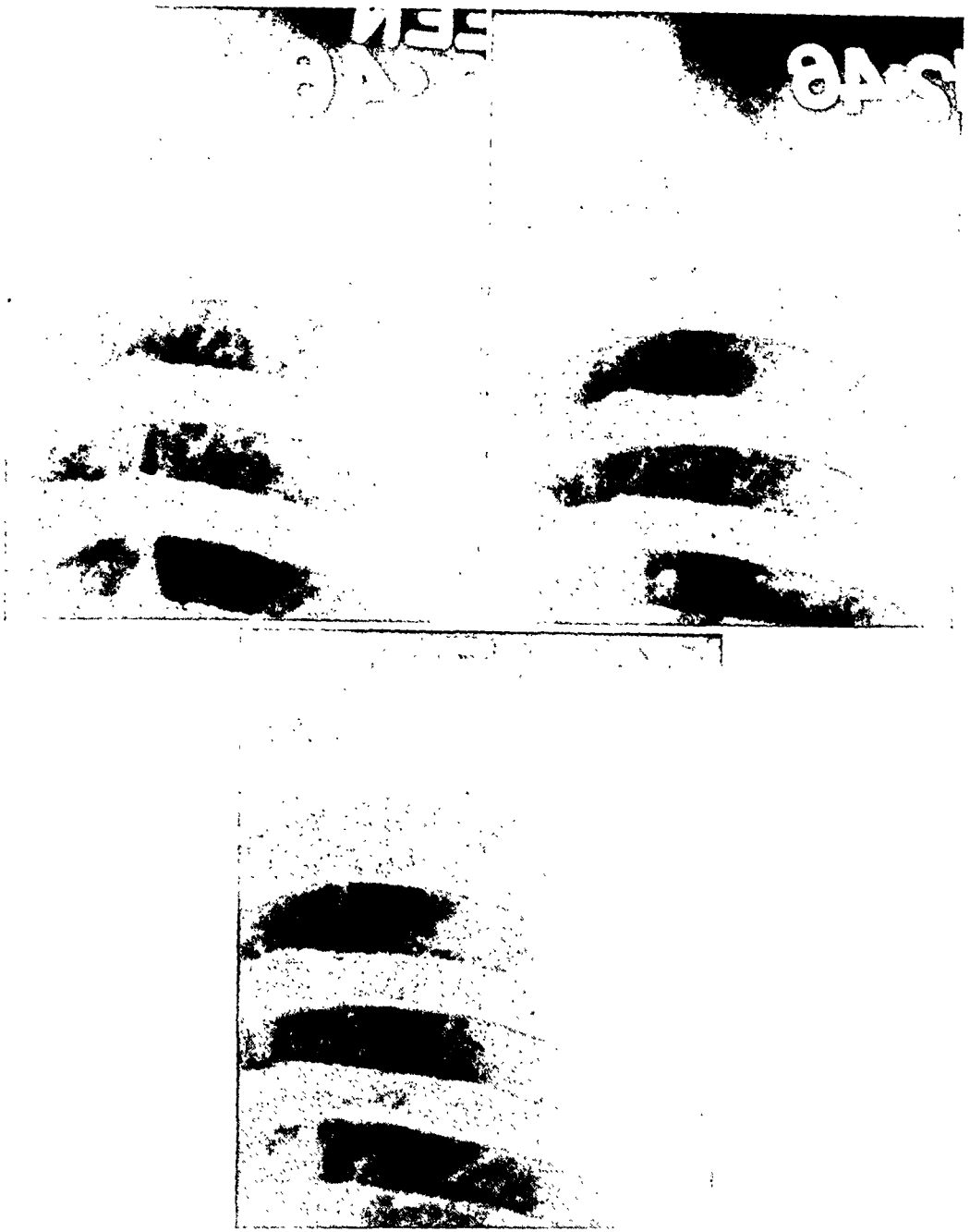
FIG. 19. (lower right) Case 5. Planigram of July 19, 1947, shows further progression of inspissation with disappearance of the central radiolucent area.

FIG. 12. (upper right) Case 4. Film of November 22, 1943. Pneumothorax has been instituted and cavity is still seen in compressed right upper lobe.

FIG. 13. (center left) Case 4. Film of Feburary 1, 1944. Cavity in right upper lobe is smaller and partly filled with secretions. Sputum negative for tubercle bacilli at this time.

FIG. 14. (center right) Case 4. Film of August 9, 1946. The nodular density in the right upper lobe represents inspissated cavity and has been present over two years.

FIG. 15. (bottom) Case 4. Film of August 22, 1947. Pneumothorax has been abandoned and inspissated cavity is still stable.



FIGS. 20-22

FIG. 20. (upper left) Case 6 (W. W.). Film of June 19, 1946, shows a small thick-walled cavity just below the left clavicle.

FIG. 21. (upper right) Case 6. Film of December 12, 1946, shows the cavity to have been replaced by a solid nodular density.

FIG. 22. (bottom) Case 6. Film of August 20, 1947. The inspissated cavity is slightly smaller and more sharply delineated.

chest roentgenograms showing a lesion typical of tuberculous cavitation, the history of recent blood spitting, daily temperature elevations, a chronic cough, and weight loss. On July 19, 1946, the tuberculin reaction was positive in the first strength using PPD. By December 12, 1946, the cavity was believed to have become completely "inspissated" (figure 21). Serial roentgenograms through August 1947 showed the lesion to have become progressively smaller (figure 22).

Comment: It is believed that the original "negative" sputum in this case can be explained by complete closure of the draining bronchus prior to the patient's hospitalization. This mechanism will be discussed later in the article.

DISCUSSION

The definition of an inspissated cavity should be thoroughly understood. It is one in which the draining bronchus has become firmly occluded, where the cavity is partially filled with secretions, and its contents have gradually thickened by the absorption of air and fluid. The finding of a horizontal fluid level within a cavity on a single chest roentgenogram is not adequate to warrant a diagnosis of permanent bronchial occlusion. On the contrary, it may indicate merely a temporary bronchial occlusion by a plug of mucus or debris which can be readily expelled. This is the so-called check-valve cavity. Only through serial films can the latter be differentiated from a cavity with a completely blocked bronchus followed by gradual absorption of air and progressive inspissation and shrinkage.

Likewise, an inspissated cavity should be differentiated from a caseous focus which has never ulcerated into a bronchus. The caseous parenchymal focus with ultimate excavation in its center is the ordinary method of cavity formation in reinfection type tuberculosis. In order to assume that a round nodular density seen on roentgenography is an inspissated cavity, there must be prior roentgenographic evidence of an open cavity in the same location.

In the past ten years a small number of articles have dealt with the mechanism of closure of tuberculous cavities. The actual anatomic evidence presented has been limited to some 39 cases (16, 17). Of these, inspissation was present in 22, being the most frequent form of healing observed in autopsy material. This seems like an extremely small number of cases when it is considered that literally hundreds of cavities have been observed in the course of collapse therapy in any large tuberculosis sanitorium. The discrepancy may be explained in two ways: 1. Those patients who collapse their cavities and "convert" their sputum are discharged and, in the main, are lost from sight and do not come to postmortem examination. 2. In those patients who die as a result of progressive pulmonary tuberculosis, what few healed cavities may be present are usually obscured by the mass of active disease. Nevertheless, several authors (Pagel (16), Auerbach and Green (12), Loesch (17)) have presented convincing anatomic evidence concerning the major mechanisms of cavity closure. These are: (*1*) closed type with inspissation or scar formation; (*2*) open type with nonspecific bronchiectatic epithelialization occurring in the cavitary wall and adjacent bronchus.

Prior to the work of these pathologists, Coryllos had adduced both theoretical

and experimental evidence to show that bronchial occlusion was of primary importance in cavity closure (1, 2, 3, 7, 8). He proved that complete closure of the bronchus through caseous bronchitis and resultant fibrosis (either in the course of bed-rest or secondary to the angulation of, and increase in pressure on, the bronchi resulting from collapse therapy) was followed by absorption of the air and shrinkage of the cavity. Coryllos clearly distinguished between this type and the temporarily blocked or check-valve cavity described by Eloesser (6) and Salkin, Cadden, and McIndoe (4). The latter will frequently balloon out and cause bronchogenic spread because of intermittent blockage by a caseous plug that subsequently may be dislodged.

In the article of Salkin *et al.* (4) which appeared in 1936, there was some confusion regarding the nature of blocked cavities. In a series of tuberculous patients who had just died they endeavored to delineate cavities and their draining bronchi. This was done with a radio-opaque material injected both into bronchi through the trachea and into cavities through the chest wall. Of 147 open cavities examined, 14 per cent did not communicate with a bronchus. Their conclusion was that blocking of a draining bronchus does not cause cavity healing.

Several fallacies should be pointed out in connection with their experiments. Certain changes occur in bronchi near or soon after the time of death. The patient has usually died in expiration (18) and lumina of the bronchi, therefore, will be narrowed. Caseous plugs, absence of respiratory movements and terminal fluid in the alveoli and bronchioles, combined with the narrowed bronchi of expiration, will cause additional bronchial stenosis. The above factors, together with the endobronchial disease known to be present near the opening of draining bronchi into cavities, may well completely occlude the bronchus. In a second article these authors admitted that blockage of bronchi often occurred due to blood, pus, secretions and entrapped air.

Although all cases of cavity closure may not be ascribed to a single mechanism according to the physiological principles enunciated by Coryllos, the draining bronchus must be of prime importance. Actually the term "inspissated cavity" is a relative one. It is highly improbable that all the caseous material is ever extruded from a cavity. With firm bronchial occlusion the retained mass is then subjected to the physiological effects of a continuing decrease in oxygen. Concomitantly with this decrease in oxygen, the metabolism of the tubercle bacillus drops. Experimentally, under such conditions the rate of growth of the bacilli may be markedly suppressed, although they may remain viable for long periods of time. Whether an inspissated cavity or a radiating scar is formed, some caseous material is always retained. Various workers have recovered viable tubercle bacilli from both types of lesions. Yet little hesitancy occurs in discharging a patient as an arrested case if a scar-type lesion has remained unchanged over a period of months. The entire concept of handling clinical tuberculosis would have to be changed if all patients were kept under hospital care until their lesions were pathologically healed. If a small mass of caseous material in the scar-type lesion is considered clinically inert, there seems to be no reason for considering a somewhat larger mass in a different light.

Whether inspissation or scar formation results may well depend upon the amount of material in the cavity at the time of bronchial occlusion, in addition to other factors such as the thickness of the cavitary wall (12). If the caseous contents have been recently extruded, the resultant mass will be smaller and eventually will contract to form a scar. If the cavity is filled at the time of bronchial closure, the resultant mass will become inspissated and remain as a round nodular density.

The idea that retention of secretions in a tuberculous cavity is harmful to a patient was based chiefly on the assumption that coexisting pyogenic organisms would cause superimposed infection. Coryllos (8), in the course of cavernostomy operations, investigated the contents of several cavities and found no pyogenic organisms on smear and culture. Even Pinner (13), who has been a leading proponent of the theory that toxemia results from retention of secretions within a cavity, has recently admitted that some cavities may follow the benign course outlined by Coryllos. The present findings in 24 cases confirm the deductions of Coryllos's physiological studies and the pathological investigations of Pagel (10), Auerbach (12) and Loesch (17).

In 20 of 24 cases the last sputum positive for tubercle bacilli was from one to sixteen months prior to the demonstration of "inspissation" by roentgenography. This is in accord with Coryllos's concept that bronchial closure precedes cavity closure. None of these patients demonstrated any clear-cut toxemic symptoms of retention.

In the past it has been the general concensus of opinion that these lesions are unsafe and eventually will empty their contents through the draining bronchus and cause a bronchogenic spread. In none of the cases of the present series has this catastrophe occurred although 8 have been followed for from two and one-half to seven and one-half years and 16 have been followed from six months to two years. Serial roentgenograms show these caseous areas to be consistently decreasing in size since "inspissation." There is good reason to believe that they will go on to calcification and permanent healing. Furthermore, in the past eighteen months at Oteen Hospital, where approximately 3,000 cases of pulmonary tuberculosis have been seen, not a single case has been observed in which a definite "inspissated" cavity has broken down. This would indicate that the latter phenomenon, although it may occur, is certainly very rare.

It should be clear that the writers are not questioning the accepted indications for pulmonary résection in certain well-defined tuberculous lesions. This procedure is generally considered to be justified in: (*1*) tuberculous bronchiectasis; (*2*) following thoracoplasty failure with a residual cavity; (*3*) basal tuberculosis which fails to respond to collapse measures; (*4*) tuberculoma. Actually the diagnosis of a tuberculoma is extremely difficult, and many round nodular lesions with an unknown diagnosis are proved to be tuberculous only following removal. Nor do the writers question the indication for exploratory thoracotomy and resection in cases of large nodular lesions of undetermined etiology (5, 11). All of the present cases involved "inspissated" tuberculous cavities which it is believed represent regressing lesions.

If, then, the inspissated cavity is accepted as being a relatively benign lesion, it ceases to be an indication for lobectomy. Examination of the tuberculous cavity at autopsy reveals that such cavities are almost never isolated lesions and caseous foci of varying size are generally scattered throughout the other lobes some of them too small to be visualized on a roentgenogram (19). It is obviously impossible to excise all the foci that may be present in a tuberculous lung. Thus it seems illogical to attempt a major surgical procedure to eradicate what is in the vast majority of cases a benign lesion, an inspissated cavity.

SUMMARY

1. The controversial subject of the fate of inspissated cavities has been discussed from the standpoint of diagnosis, pathogenesis and prognosis.

2. Twenty-four cases are presented in which presumably inspissated cavities have been followed for as long as seven and one-half years. Histories on six of these patients are given in detail, together with serial roentgenograms showing the course of inspissation.

3. It is concluded that a true inspissated cavity is essentially a benign lesion and has a favorable prognosis.

SUMARIO

Pronóstico de las Cavernas Espesadas

1. Discútese, desde el punto de vista del diagnóstico, patogenia y pronóstico, el debatido tema del destino de las cavernas espesadas.

2. Preséntanse 24 casos en los que cavernas presuntamente espesadas fueron observadas hasta siete años y medio. Se dan con todo detalle las historias clínicas de seis de esos enfermos, junto con radiografías seriadas que revelan la evolución del espesamiento.

3. Dedúcese que una verdadera caverna espesada es en el fondo una lesión benigna con un pronóstico favorable.

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THE EFFECT OF NONTUBERCULOUS PULMONARY INFLAMMATION ON PULMONARY TUBERCULOSIS[1,2]

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INTRODUCTION

The following paper is an outgrowth of a study on the same subject (1), published in 1942 by Dr. J. Burns Amberson and one of the writers (O. S. B.), in which the effects of nontuberculous inflammation on pulmonary tuberculosis were observed. An apparent increase in the incidence of coexistent disease and a new therapeutic approach to the pneumonias has prompted an additional review of the subject.

The primary purpose of this paper is to determine: (*a*) the actual incidence of complicating nontuberculous inflammation; and (*b*) the effect of chemotherapy on the course of the underlying tuberculous process.

A brief resumé of the conclusions of the original report is in order. It was found that pneumonia in the tuberculous population was not as rare as formerly had been thought, that the necrotizing process[3] within a pneumonic area was the most important factor in activation of a tuberculous lesion located therein; and that the necrotizing process usually affected the tuberculous lesion adversely. Interstitial extension of the infecting organism from the alveoli, such as occurs in the pneumonias produced by type III *pneumococcus*, *hemolytic streptococcus*, *staphylococcus*, and *Friedländer's bacillus*, was also found to be one of the determining factors. The destructive change is also probably related to local toxins and the action of enzymes. Bronchorrhea and local hyperaemia may also play a role, particularly when the tuberculous lesion is located at a distance from the superinfecting process. Fibrocalcific lesions and lesions successfully controlled by collapse therapy were most resistant to breakdown.

Since 1942 there has been only one publication on this subject, that of Hogan (2) who studied 111 cases of pneumonia occurring in tuberculous individuals admitted over an eight year period to the Philadelphia General Hospital. It was his impression that the incidence of coexisting lesions was relatively low, and that pneumonia occurred primarily as a terminal event. In 15 per cent of his series he found evidence of activation of a tuberculous focus. This occurred whether the tuberculosis had previously been quiescent or active, whether or not it was situated in the same lobe with the pneumonia, and whether or not the latter was specifically treated. This study was made prior to the advent of penicillin

[1] From the Chest Service, Bellevue Hospital, New York, and Department of Medicine, College of Physicians and Surgeons, Columbia University, New York.

[2] Presented at the annual meeting of the New Jersey State Medical Society, April 1948.

[3] Much confusion exists in the literature concerning the terminology of the various types of pneumonia. The term "suppurative pneumonia" has frequently been employed to denote the presence of a necrotizing component of the disease process. In this paper the term "necrotizing pneumonia" is used to differentiate this process from purely intra-alveolar pneumonias and from interstitial (atypical, "virus") pneumonias.

therapy. It is of importance to note that the pneumonias in his study were primarily nonsuppurative. Moreover, his concern was mainly with the effect of tuberculosis on the complicating pneumonia in contrast to the approach of the present study.

CLINICAL MATERIAL

The material comprising the present study consists of all cases of proven coexisting tuberculosis and pneumonia, necrotizing and otherwise, discharged from the chest service of Bellevue Hospital between July 1941 and June 1947. All cases of tuberculous pneumonia, pneumonia secondary to tuberculous, or neoplastic endobronchial disease are excluded.

Evidence of activation of the tuberculous lesion was considered to be *definite* when there was: (*a*) post mortem evidence of recent activation; (*b*) progressive tuberculosis after pneumonia, and the tuberculosis had previously been stationary; (*c*) extension of previously active tuberculosis coincidentally with the pneumonia.

TABLE 1

Incidence of activation

PNEUMONIA			NECROTIZING PNEUMONIA		
Activation	Number	Per cent	Activation	Number	Per cent
Definite	16	13	Definite	21	55
Doubtful	9	8	Doubtful	4	11
No activation	91	78	None	11	29
Undetermined	1		Undetermined	2	5
Total	117	100	Total	38	100

Cases in which the pneumonia was accompanied or followed by the transient expectoration of tubercle bacilli or residual densities on the roentgenogram were classified as *doubtful*. The designation *undetermined* was used for those cases in which death occurred too quickly for proper evaluation or in which there was inadequate information regarding pre-existing tuberculosis.

A total of 155 cases, 38 of which were necrotizing pneumonias, has been reviewed. In table 1 the criteria of activation are applied separately to the pneumonias and necrotizing pneumonias. The high percentage (55 per cent) of activation in the necrotizing group is of interest.

The relationship between activation and the type of pre-existing tuberculous disease may be seen in table 2. The largest percentage of activation (11 per cent) is seen to occur in the "fibro-caseous" classification.

As was pointed out in the previous report, the bacteriology and location of the inflammatory lesion are of greatest importance in determining subsequent activation of the tuberculosis. In tables 3 and 4 may be seen the relationship of activation to these factors. It may be seen that organisms causing destruction of lung tissue are most likely to cause activation when the lesion produced

thereby is located in a tuberculous lobe. In contrast, the less destructive types of bacteria, especially when situated in a distant lobe, rarely cause activation.

TABLE 2

Relation between activation and type of disease (necrotizing pneumonias excluded)

ACTIVATION	TYPE OF TUBERCULOSIS					
	Fibro calcific		Fibro-caseous		Exudative	
	Number	Per cent	Number	Per cent	Number	Per cent
Definite	2	2	13	11	1	1
Doubtful	2	2	5	4	2	2
No activation	56	47	25	21	10	9
Undetermined			1	1		

TABLE 3

Relation between activation and bacteriologic findings

	Pneumococci				*Streptococci* OR *Staphylococci hemolyticus*	*K. pneumoniae*	MIXED AND UNDETERMINED
	I	II	III	Other *Pneumococci*			
Definite	1	3	1	9	4	4	16
Doubtful	0	0	1	6	1	0	5
No activation	5	9	4	21	2	1	59
Undetermined	0	0	0	0	1	0	2
Total	6	12	6	36	8	5	82

TABLE 4

Relation between activation and location

ACTIVATION	LOCATION OF TUBERCULOUS LESION AND NONTUBERCULOUS PNEUMONIA								
	Same lobe			Other lobes			Undetermined		
	P	N	T	P	N	T	P	N	T
Definite	13	14	27	1	7	8	2	0	2
Doubtful	3	4	7	5	0	5	1	0	1
No activation	18	0	18	73	11	84	0	0	0
Undetermined	1	2	3	0	0	0	0	0	0
Total			55			97			3

P = Pneumonia.
N = Necrotizing pneumonia.
T = Total.

Activation of the tuberculous process occurred in only 2 of 6 cases of *pneumococcus* type III pneumonia (table 3). This apparent contradiction to the findings in the previous paper of the very invasive nature of this organism is clarified

by referring to table 5 in which the relationship between activation, etiology of the acute pneumonia and its location are tabulated. It may be seen that 3 of the remaining 4 cases of *pneumococcus* type III pneumonia occurred in lobes other than those involved by tuberculosis. It is of interest to note that the fourth case of nonactivation occurred in a tuberculous lobe which was effectively collapsed by pneumothorax. In table 6 the relation of therapy to activation is tabulated. In this small number of cases chemotherapy appeared to have no effect on the percentage of activation.

TABLE 5

Relation between bacteriologic findings and location of the pneumonia to activation of tuberculosis

ACTIVATION	*Pneumococci*								*Streptococci* OR *Staphylococci hemolyticus*		*K. pneumoniae*		MIXED AND UNDETERMINED	
	I		II		III		Other *Pneumococci*							
	S	O	S	O	S	O	S	O	S	O	S	O	S	O
Definite	1	0	3	0	1	0	7	2	3	1	4	0	9	7
Doubtful	0	0	0	0	0	1	3	3	1	0	0	0	3	2
No activation	0	5	2	7	1	3	5	16	0	2	0	1	9	50
Undetermined	0	0	0	0	0	0	0	0	1	0	0	0	2	0
Total	6		12		6		36		8		5		82	

S = Same lobe. O = Other lobes.

TABLE 6

Relation of therapy to activation

	PNEUMONIA				NECROTIZING PNEUMONIA			
	Definite	Doubtful	No activation	Undetermined	Definite	Doubtful	No activation	Undetermined
Sulfonamides	5	5	46	0	5	2	2	1
Penicillin	9	3	12	0	7	0	1	1
Combined therapy	2	1	3	0	2	0	1	0
No specific therapy	0	0	30	1	7	2	7	0
Total	16	9	91	1	21	4	11	2

CASE REPORTS

Case 1: S. M., a 52-year-old white male, was admitted February 13, 1945 with one week's history of left chest pain, cough and rusty sputum. The patient was acutely ill, with signs of left lower lobe pneumonia. The total leucocyte count was 15,000 per cu. mm. The sputum contained type I *pneumococci* and was persistently negative for acid-fast bacilli. The patient was treated with sulfadiazine, with a dramatic clinical response but a slow resolution of pneumonia on the roentgenograms. There was no activation of an old left upper lobe infiltrate.

Case 2: J. K., a 61-year-old white male, was admitted to another hospital five days after onset of chills, fever, and cough. Left lower lobe pneumonia was diagnosed and treatment with sulfathiazole was started. Two days later, on January 27, 1944, he was transferred to the Psychiatric Division of Bellevue Hospital because of psychotic symptoms. On admission there were signs of a left lower lobe pneumonia and a leucocytosis of 11,000 cells per cu. mm. Results of bacteriologic studies of the sputum are unknown except for persistent absence of acid-fast bacilli. All symptoms subsided rapidly. The pneumonia resolved slowly. There was no activation of an old tuberculosis of the left and right upper lobes.

Case 3: W. A., a 21-year-old white male, had known tuberculosis since 1941 which had been treated by right pneumothorax since March 1942. He was admitted on July 15, 1943 with a four day history of fever, chills, cough, sputum, and chest pain. On admission he was acutely ill. The total leucocyte count was 11,000 per cu. mm. with 70 per cent polymorphonuclears. The sputum contained type III *pneumococci* and was persistently negative for acid-fast bacilli. A roentgenogram revealed pneumonia in the collapsed lung. The clinical symptoms subsided rapidly on sulfadiazine therapy and the roentgenographic findings reverted to the pre-pneumonia status. There was no activation of the tuberculous lesions.

Case 4: C. V., a 50-year-old white male, with known tuberculosis since 1938, treated by thoracoplasty in March 1942, was admitted on April 27, 1943 with a two day history of cough, sputum, fever, chills, and right chest pain. The temperature was 104° F. and the total leucocyte count was 11,000 per cu. mm. The sputum contained type XIX *pneumococci* and was persistently negative for acid-fast bacilli. Roentgenographic examination revealed a density in the collapsed right lung. The patient was treated with sulfadiazine with dramatic response although leukopenia developed on the third day and administration of the drug was stopped. There was progressive clearing of the density visible on the roentgenogram. The patient has been followed to date and no evidence of activation of his tuberculosis has appeared.

Case 5: P. L., a 46-year-old white male with known minimal right upper lobe tuberculosis since 1945 which had been treated by bed-rest in a sanatorium for one year, was admitted on April 7, 1947 with a two week history of upper respiratory infection and a one day history of right pleuritic pain and fever. He was moderately ill and had a leucocytosis of 18,000 cells per cu. mm. with 86 per cent polymorphonuclears. A chest roentgenogram revealed no extension of the minimal tuberculosis. The acute pneumonia was located in the apex of the right lower lobe. Culture of the sputum showed *streptococcus hemolyticus* and *streptococcus viridans, staphylococcus albus,* and *M. catarrhalis.* The patient was treated with penicillin with prompt clinical and roentgenological response. Unfortunately, however, there was a progressive increase in the formerly inactive tuberculous lesion. This case represents an instance of definite activation following an acute non-tuberculous pneumonia.

Case 6: P. P., a 45-year-old white male, had known tuberculosis of the right upper lobe since 1938 with subsequent spread to the left mid-lung. A cavity had been present in the right apex until 1940 when it closed. He was admitted November 4, 1942 with a short history of fever, cough and chest pain. There were signs of consolidation in the left lower lobe and type VIII *pneumococci* were found in the sputum. There was a prompt clinical response to sulfadiazine with complete clearing of the pneumonia and no acti-

vation of the tuberculosis. He was readmitted in October 1945 with a one week history of chills, fever, cough, sputum, and sudden left chest pain. On admission he was acutely ill, dyspneic, and cyanotic. The total leucocyte count was 20,000 per cu. mm. A roentgenogram of the chest showed a left lower lobe pneumonia with a left spontaneous pneumothorax. Sputum culture showed *streptococcus hemolyticus*, *streptococcus viridans*, and *E. coli*. The patient was treated with penicillin with complete resolution of the pneumonia and re-expansion of the left lung. The sputum was persistently negative for acid-fast bacilli. He was discharged in December 1945, but readmitted in February 1946 with a history of progressive cough, sputum and weight loss since discharge. The sputum contained acid-fast bacilli. A chest roentgenogram showed extensive left upper lobe disease with cavitation and subsequent marked spread. This case represents an instance of definite activation of tuberculosis following an acute nontuberculous pneumonia.

Case 7: D. R., a 68-year-old white male, was perfectly well until four days before admission in May 1941 when he began to have left chest pain and a productive cough. He was acutely ill with signs of left upper lobe lobar and scattered bronchopneumonia. The total leucocyte count was 22,000 per cu. mm. with 83 per cent polymorphonuclears. The sputum contained type III *pneumococci* and on 12 examinations was negative for acid-fast bacilli. The patient was treated with sulfadiazine and 175,000 units of serum with good clinical improvement, and he was discharged July 7, 1941. He was readmitted on September 5, 1941 because of cough beginning one month after discharge. The roentgenogram revealed an infiltration of the left upper lobe and throughout the right lung. The sputum was persistently positive for acid-fast bacilli. This case is considered to represent a definite activation following an acute nontuberculous pneumonia.

Case 8: D. G., a 41-year-old white male, was admitted on April 9, 1945 with a history of an alcoholic debauch on the previous New Year's Eve, followed by the aspiration of vomitus. In February he began to cough, producing one-half cup of mucopurulent, occasionally foul, sputum and complained of weakness and a 20 pound weight loss. Examination revealed a temperature of 100° F., foul gums, and rales over the right upper lobe. The first five sputum examinations were negative for acid-fast bacilli. Successively thereafter one was positive, nine were negative and two were positive. Subsequently the sputum examinations were intermittently "positive" from May until July. The total leucocyte count ranged between 7,000 and 14,000 cells per cu. mm. with 80 to 87 per cent polymorphonuclears. Cultures of the sputum revealed the usual mouth flora. On bedrest the patient gained weight and the lesion cleared rapidly. Bronchoscopy and bronchography were negative. It was felt that this was a necrotizing pneumonia with activation of a tuberculous focus.

Case 9: M. C., a 43-year-old white male alcoholic, was admitted November 21, 1941 with a one week history of cough, sputum, and fever. He was found to have a right upper lobe pneumonia and *K. pneumoniae*, type B, was cultured from sputum and blood. He was treated with sulfapyridine. He showed a slow clinical response with gradual excavation of upper lobe followed by a residual abscess. The patient refused permission for pulmonary resection.

He was followed for one and one-half years and no change was noted on the roentgenograms. Nevertheless, he had repeated acute respiratory episodes with several positive cultures for *K. Pneumoniae*, type B. Examination of the sputum was always negative for acid-fast bacilli.

He was readmitted December 6, 1943, having been lost from observation during the

preceding six months. On this admission he gave a history of cough, sputum and 36 pound weight loss in the preceding two months. Roentgenographic examination revealed extensive bilateral infiltration and the sputum was positive for acid-fast bacilli.

DISCUSSION

From the data presented it is evident that the incidence of the diagnosis of coexisting pulmonary diseases on the Chest Service of Bellevue Hospital has markedly increased. It is believed that this is due to an increased awareness of the possibility of such coexistence rather than an increase of pneumonia or tuberculosis, or of pneumonia occurring in the tuberculous. It is the impression of the writers that many cases of tuberculosis are overlooked when admitted to a general hospital with the diagnosis of pneumonia. One need only inquire into the past history of the average tuberculous individual to observe the great frequency of "pneumonia," with classical symptoms and findings, rapid subsidence of these symptoms with appropriate therapy, early discharge from the hospital, and subsequent readmission for active tuberculous disease. Obviously these cases cannot be included in such a study as the present one. It seems certain, however, that their number is great and that the actual incidence of coexisting pulmonary diseases is larger than any available statistics show. It is believed that a true picture can be obtained by attention to the following principles: (*1*) roentgenographic examination of all cases of pneumonia just before discharge from the hospital; (*2*) careful sputum studies in all suspicious cases; (*3*) close observation of all cases showing slow resolution of an acute pneumonia; and (*4*) particular attention to those cases where the course and bacteriological findings suggest the possibility of a necrotizing process.

The present data reinforce the concept presented previously that a tuberculous lesion is most likely to be activated by a pneumonia if the latter is necrotizing and is located in the same lobe. Although the greatest incidence of activation is in active fibro-caseous disease, the nature of the original tuberculous lesion is of relatively minor importance, as activation can occur in calcified primary lesions.

Despite the relatively small number in this series, the sulfonamide and penicillin treated cases show a rather high incidence of activation. Comparison with previous statistics shows that use of chemotherapy has not significantly changed the percentage of activation. One may speculate as to the probable reasons for this disappointing observation. Is it justified to assume that specific therapy of the pneumonia prolongs life and hence affords time for the progression of activated tuberculous disease? Or is it possible that in nonterminal cases the early invasive nature of the superinfecting organism initiates progression of the preexisting tuberculous lesion before chemotherapy has a chance to exert its bacteriostatic or bactericidal effect? Theoretically it seems likely that intensive specific therapy, if instituted early, would prevent the development of necrosis and thereby prevent activation of the underlying tuberculous disease. The disappointing results might thus be due to late initiation of therapy, a situation which might be remedied by the admission of patients early in the course of their pneumonia. Whether or not coincident administration of streptomycin

would prevent activation of the tuberculous process remains to be determined in further observations.

SUMMARY

1. A study of the effect of nontuberculous inflammation on pulmonary tuberculosis has been brought up to date. The increased incidence of coexisting disease is probably due to increasing awareness of the possibility.

2. The necrotizing component of a pneumonia is again shown to be of greatest significance in the activation of tuberculous infection. Location of the superinfection is again implicated.

3. Satisfactory collapse therapy militates against activation even when pneumonia is located in the collapsed lobe.

4. Adequate bacteriological identification of the superinfecting organism is essential to the proper management of the tuberculous disease and has a direct bearing on prognosis.

5. The present limited data indicate that antimicrobial therapy did not prevent activation of the tuberculous process. On theoretical grounds early intensive specific therapy might change these results.

SUMARIO

Efecto de la Neumonitis No Tuberculosa sobre la Tuberculosis Pulmonar

1. Se ha puesto al día un estudio del efecto de la inflamación no tuberculosa sobre la tuberculosis pulmonar. La mayor incidencia de esta coexistencia procede probablemente de la mayor atención prestada a su posibilidad.

2. Demuéstrase de nuevo que el factor necrotizante de una neumonía reviste importancia máxima en la activación de la infección tuberculosa. La localización de la superinfección es inculpada de nuevo.

3. La colapsoterapia satisfactoria milita contra la activación aun cuando la neumonía se localice en el lóbulo aplastado.

4. La adecuada identificación bacteriológica del microbio superinfectante resulta indispensable para la propia atención de la enfermedad tuberculosa y guarda relación directa con el pronóstico.

5. Los actuales y limitados datos indican que la terapéutica antimicrobiana no impidió la activación del proceso tuberculoso. Teóricamente, una intensa y temprana terapéutica específica podría alterar este resultado.

Acknowledgment

The authors wish to express their thanks to Dr. J. Burns Amberson for his invaluable assistance in the preparation of this paper.

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A SUPERNUMERARY RIB[1]

SYDNEY JACOBS

REPORT OF A CASE

B. T., a 43-year-old Negro male, was admitted to the Charity Hospital of Louisiana in May, 1946, because of bilateral far advanced pulmonary tuberculosis. On admission,

FIG. 1. Chest roentgenogram revealing supernumerary rib on the right.

there was roentgenographic evidence (figure 1) of a supernumerary rib. The patient died on June 27, 1946. At autopsy the most important findings were bilateral caseous pneu-

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monia with extensive cavitation, miliary tuberculosis of the spleen and kidneys, and syphilitic aortitis. No congenital anomalies other than the supernumerary rib were found.

When the right lung was removed, the supernumerary rib was seen originating from the third dorsal vertebra on its anterolateral surface, articulating independently of the normal rib arising from this vertebra. The rib coursed along the posterior thoracic wall downward and slightly laterally on the anterior surface of the normal ribs, external to the pleura. It terminated at the level of the eighth thoracic vertebra seven centimeters from the vertebral body. There were twelve pairs of thoracic ribs arising in normal manner.

About one per cent of persons have some variation from the normal pattern of twelve pairs of thoracic ribs. Extra ribs, in either the cervical or the lumbar region, may be single or paired. Occasionally one side of the thorax may have fewer than twelve ribs because of maldevelopment of a vertebra (1) or because of fusion of adjacent vertebral or costal segments. Such variations can easily be explained and are usually accompanied by other congenital anomalies.

The herein reported supernumerary rib cannot be similarly explained and was not found associated with other congenital anomalies. No description of a similar rib could be found in the literature.

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A TUBERCULOSIS CASE-FINDING PROGRAM IN ERIE COUNTY, NEW YORK

VINCENT H. HANDY[1] AND WILLIAM D. CRAGE[2]

INTRODUCTION

For many years Erie County, including the city of Buffalo, has had one of the highest tuberculosis death rates in New York State. During the period 1926–1928 the average annual number of tuberculosis deaths among residents was 561, giving a mortality rate of 78.0 per 100,000 population. In 1944–1946 the average annual number of deaths among residents was 426; this figure gives a rate of 49.6 per 100,000 (1).

The tuberculosis death rate for Erie County in 1946 was 46.8 per 100,000 population. Although this rate is considerably lower than those recorded twenty years ago, it is obvious that Erie County still has excessively high mortality when one considers that in 1946 the tuberculosis death rate for the entire upstate New York area was 30.2 per 100,000 (1). With such a high death rate one might reasonably expect a high incidence of the disease. This assumption led to an examination of the local facilities available for tuberculosis case-finding in 1946.

The county has two health department clinics where individuals may obtain roentgenograms without charge. One is operated by the Buffalo City Health Department and one by the Erie County Health Service. In addition to these clinics, the city of Buffalo has several approved hospitals where chest clinics are held and where chest roentgenograms are obtainable. A chest clinic is also operated by the Buffalo and Erie County Tuberculosis Association.

In spite of these facilities, however, the city had had no concentrated drive to get the population examined roentgenographically and to find as many cases as possible, although the Tuberculosis Association had conducted several mass projects embracing some 10,000 roentgenograms over a period of three years.

The nursing services of the official health agencies did the follow-up work in connection with these projects. It became obvious, however, that more positive measures would have to be taken in order to find the unknown cases.

After study, the Board of Directors of the Buffalo and Erie County Tuberculosis Association purchased a mobile roentgenographic unit from Christmas Seal funds. On March 4, 1946, a mass tuberculosis case-finding program was undertaken for Buffalo and all of Erie County by the Tuberculosis Association. This program had the support of the Erie County Medical Society, the state and local health departments, and various other health agencies in the community.

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PLAN OF STUDY

At the beginning of the program the goal was to examine roentgenographically approximately 60,000 persons during the year (March 1946–March 1947). Actually this aim was attained in December 1946. By the end of March 1947, a period of thirteen months, 81,253 roentgenograms had been taken, an average of more than 6,000 per month. This survey was carried on in the city of Buffalo and the remainder of Erie County except for the city of Lackawanna, where a mass roentgenographic project was actively begun in April 1947.

The persons examined were distributed in all age groups and in a wide variety of occupational pursuits. As the total population of Erie County, including Buffalo, was estimated to be 869,571 on July 1, 1946 (1), it is evident that 9.3 per cent of the county's population had been examined roentgenographically by the Christmas Seal mobile unit during its first thirteen months. If only the adult population is considered, then approximately 11.1 per cent of the residents 15 years and over were examined during the thirteen month period.

Of the 81,253 roentgenograms taken, 741 proved to be unsatisfactory, duplicates, or test films. Accordingly, a total of 80,512 persons examined is used as the basis of this study.

The equipment used was a Picker unit in a tractor-trailer combination taking a 70 mm. film. All the films were developed at the clinic of the Tuberculosis Association and were read by a physician of its staff. When a tentative diagnosis of tuberculosis was made, it was based on the characteristic changes in density on the roentgenogram which are presumed to be tuberculous. Persons whose films indicated the need of further study were sent appointment letters within ten days which arranged for first follow-up examination at the office of the Association. These examinations included 14 by 17 inch roentgenograms.

The purpose of the present article is to analyze the 80,512 persons examined with 70 mm. "miniature" film during the thirteen month interval from March 4, 1946, to March 31, 1947, and to ascertain what has happened to those requested to return for follow-up examination.

OBSERVATIONS

In table 1 may be seen the total number of men and women examined by miniature film during the period studied, classified by age group.

Nearly 7 per cent of the persons examined roentgenographically were less than 15 years of age; this group is generally recognized to be unproductive so far as tuberculosis case-finding is concerned. Roentgenograms were obtained of children 12 to 14 years old only if accompanied by their parents, while those under 12 who were contacts were also examined. Subsequent tables show that very little indication of pathology appeared on the miniature films taken of children in this group.

Table 2 gives the distribution of persons examined by race, sex, and age. Since but 3,504 nonwhite persons were examined, the number was considered too small for detailed analysis.

According to table 3, 190 of the 80,512 persons examined were given preliminary diagnoses of definite tuberculosis on the miniature film, while 971 others were suspected of having the disease. For each 1,000 persons examined, 2.4 tentatively definite cases were found in addition to 12.1 suspected cases.

TABLE 1

Persons of each sex who were examined by miniature film, classified by age group: Erie County, March 4, 1946–March 31, 1947

AGE GROUP	BOTH SEXES		MALE		FEMALE	
	Number	Per cent distribution	Number	Per cent distribution	Number	Per cent distribution
All ages	80,512	100.0	38,711	100.0	41,801	100.0
Under 15 years	5,464	6.8	2,496	6.4	2,968	7.1
15 to 24 years	29,164	36.2	13,527	34.9	15,637	37.4
25 to 34 years	16,176	20.1	7,390	19.1	8,786	21.0
35 to 44 years	13,808	17.2	6,651	17.2	7,157	17.1
45 to 54 years	9,252	11.5	4,852	12.5	4,400	10.5
55 to 64 years	4,673	5.8	2,630	6.8	2,043	4.9
65 years and over	1,483	1.8	899	2.3	584	1.4
Age not reported	492	0.6	266	0.7	226	0.5

TABLE 2

White and nonwhite persons of each sex who were examined by miniature film, classified by age group: Erie County, March 4, 1946—March 31, 1947

AGE GROUP	WHITE			NONWHITE		
	Both sexes	Male	Female	Both sexes	Male	Female
All ages	77,008	37,191	39,817	3,504	1,520	1,984
Under 15 years	5,184	2,356	2,828	280	140	140
15 to 24 years	28,042	13,068	14,974	1,122	459	663
25 to 34 years	15,284	7,062	8,222	892	328	564
35 to 44 years	13,206	6,376	6,830	602	275	327
45 to 54 years	8,850	4,652	4,198	402	200	202
55 to 64 years	4,539	2,548	1,991	134	82	52
65 years and over	1,432	876	556	51	23	28
Age not reported	471	253	218	21	13	8

The interesting feature of table 3 is the fact that the proportion of cases found increases directly with the age of those examined. This statement holds whether one considers the proportion of persons with tentative diagnoses of definite tuberculosis or the proportion with suspected tuberculosis.

This table corroborates the findings of certain other studies. The success of a mass case-finding project hinges on ability to induce large numbers of older persons to be examined, though this fact is not widely recognized. The higher

the median age of the group examined, the larger will be the number of cases found.

Among the white persons examined, 2.4 per 1,000 were tentatively diagnosed as having definite tuberculosis; this proportion is the same as that for the entire group. The corresponding proportion for the nonwhite group was 1.7 per 1,000 on whom roentgenograms were obtained.

Cases considered suspicious among the white group numbered 11.9 per 1,000 examined, compared to 14.8 per 1,000 nonwhite persons.

Roentgenograms were obtained of a larger proportion of the Buffalo residents than in the remainder of the county, but the proportion of definite and suspicious cases found was slightly higher in the county.

TABLE 3

Persons examined by miniature film and number and per cent with tentative diagnoses of definite or suspicious tuberculosis, classified by age group: Erie County, March 4, 1946–March 31, 1947

AGE GROUP	PERSONS EXAMINED ROENTGENOGRAPHICALLY	PERSONS WITH TENTATIVE DIAGNOSES OF DEFINITE TUBERCULOSIS		PERSONS WITH SUSPICIOUS TUBERCULOSIS	
		Number	Rate per 1,000 persons examined	Number	Rate per 1,000 persons examined
All ages	80,512	190	2.4	971	12.1
Under 15 years	5,464	1	0.2	8	1.5
15 to 24 years	29,164	23	0.8	144	4.9
25 to 34 years	16,176	30	1.9	180	11.1
35 to 44 years	13,808	44	3.2	216	15.6
45 to 54 years	9,252	42	4.5	203	22.0
55 to 64 years	4,673	37	7.9	143	30.6
65 years and over	1,483	13	8.8	73	49.2
Age not reported	492	—	—	4	8.1

Although tentative diagnoses were made on the basis of miniature films, more definite diagnoses occurred after a follow-up examination and study. In the miniature films, tentative diagnoses of so-called definite tuberculosis were made in 190 cases, suspect tuberculosis in 971, and nontuberculous abnormalities in 383 cases. A total of 1,576 persons needing examination and second roentgenograms were invited to return to the chest clinic of the Buffalo and Erie County Tuberculosis Association. In response to this request, 1,490 persons were examined; 1,298 of these returned to the Association clinic for their first follow-up examinations and 14 by 17 films. This number includes 147 persons tentatively diagnosed as having definite tuberculosis, 811 with suspect tuberculosis, 308 with nontuberculous abnormalities, and 32 whose miniature films were unsatisfactory.

The names of those who failed to appear for this follow-up examination at the Tuberculosis Association's clinic were turned over to the health departments for

subsequent follow-up. This group was referred either to the City Chest Clinic or to the Erie County Health Service, depending upon their places of residence. There were 192 of this group examined, including 29 who were tentatively diagnosed as definite tuberculosis on the miniature film, 108 considered to be tuberculosis suspects, and 55 with nontuberculous abnormalities.

In table 4 are presented the findings on the follow-up of 1,490 persons on whom 14 by 17 films were taken at these three clinics. According to this table, a total of 335 clinically significant cases of tuberculosis was found as a result of examining 80,512 persons by roentgenogram. These figures indicate that 4.16 clinically significant cases were found per 1,000 examined. This finding is

TABLE 4

Cases diagnosed on 14 by 17 film, classified by tentative diagnosis on miniature film: Erie County, March 4, 1946–March 31, 1947

TENTATIVE DIAGNOSIS ON MINIATURE FILM	TOTAL EXAMINED BY 14 BY 17 FILM	CLINICALLY SIGNIFICANT CASES OF PULMONARY TUBERCULOSIS DIAGNOSED BY 14 BY 17 FILM				SUSPICIOUS TUBERCULOSIS	APPARENTLY CURED TUBERCULOSIS	NONTUBERCULOUS ABNORMALITIES	ESSENTIALLY NEGATIVE	EXAMINATION PENDING
		Total	Minimal	Moderately advanced	Far advanced					
All diagnoses	1,490	335	255	73	7	68	242	388	423	34
"Definitely tuberculosis"	176	118	66	45	7	5	34	10	2	7
Suspicious tuberculosis	919	212	186	26	—	55	190	128	316	18
Nontuberculous abnormalities	363	3	2	1	—	8	17	247	79	9
Cardiac condition	21	—	—	—	—	—	—	19	2	—
Suspected silicosis	16	—	—	—	—	1	—	13	1	1
Other pathology	326	3	2	1	—	7	17	215	76	8
Unsatisfactory film	32	2	1	1	—	—	1	3	26	—

comparable to that of other mass case-finding projects in which approximately 4 active cases were found per 1,000 *adults* examined.

A check of health department records revealed that 23, or approximately 7 per cent, of these 335 clinically significant cases were previously known. As stated earlier in this article, the number of cases found depends largely upon the ages of the persons examined.

In addition to the clinically significant cases of tuberculosis, 68 cases were considered to be suspicious tuberculosis, 242 were found to be apparently cured, and 388 individuals showed evidence of some nontuberculous abnormality. The last named group included instances of marked fibrosis, emphysema, pneumonia, neoplasm, or cardiac enlargment which would warrant further study.

Of the 176 cases diagnosed as "definite tuberculosis" on the miniature films two-thirds (118) were considered to be clinically significant when the 14 by 17 films were scrutinized. Moreover, 23 per cent of the 919 persons whose miniature films implied suspicious tuberculosis proved to have the disease in a clinically significant stage when the larger films had been taken.

Three-fourths (255) of the 335 clinically significant cases found in this survey were diagnosed as in the minimal stage. This percentage compares favorably with the findings of other mass case-finding projects.

A summary of the significant findings among the 80,512 persons examined is shown in table 5. It is interesting to note that the tuberculosis cases found are less numerous than are the cases with nontuberculous abnormalities.

From the data in table 5 it is evident that 1.28 per cent of the 80,512 persons examined were found to have some stage of tuberculosis or some other pathological condition. If the apparently cured group is disregarded, then one per cent of those examined roentgenographically needed further supervision. A report was sent in each instance to the patient's private physician.

TABLE 5

Number and rate of pathological cases found per 1,000 examined, classified by diagnosis on 14 by 17 film: Erie County, March 4, 1946–March 31, 1947

DIAGNOSIS ON 14 BY 17 FILMS	PATHOLOGICAL CASES FOUND	
	Number	Per 1,000 persons examined
All diagnoses	1,033	12.83
Clinically significant tuberculosis	335	4.16
Suspected tuberculosis	68	0.84
Apparently cured tuberculosis	242	3.01
Nontuberculous abnormalities	388	4.81

TABLE 6

Persons with specified diagnoses on miniature films who had failed to return for follow-up by April 1, 1947, classified by reason for failure to appear: Erie County, March 4, 1946–March 31, 1947

REASON FOR FAILURE TO APPEAR FOR FOLLOW-UP	PERSONS WITH SPECIFIED DIAGNOSES ON MINIATURE FILM WHO FAILED TO RETURN FOR FOLLOW-UP			
	Total	"Definite tuberculosis"	Suspicious tuberculosis	Nontuberculous abnormalities
All reasons	86	14	52	20
Refused to be re-examined	47	5	29	13
Under care of private physician	20	7	12	1
Could not be located	17	1	11	5
Returned to tuberculosis sanatorium	2	1	—	1

In surveys of this size it is impossible to obtain 100 per cent follow-up. For example, 86 persons failed to appear who were supposed to report for follow-up after their miniature films had been read. The names of these persons were referred to the city health department and were checked by public health nurses.

In table 6 may be seen the reasons for failure to return for follow-up examinations, together with the initial diagnoses on the miniature films. As 1,576 were requested to return and only 47 definitely refused to do so, it is evident that the

uncooperative group was but 3 per cent of those who needed follow-up. It is difficult to determine whether the group which could not be located were uncooperative or not.

SUMMARY AND CONCLUSIONS

Between March 4, 1946 and March 31, 1947, a mass case-finding project was carried on in Erie County, New York by the Buffalo and Erie County Tuberculosis Association. During this period 80,512 persons had chest roentgenograms (miniature films) and 1,576 were found to need further study. The first step in this follow-up procedure consisted of examination and another roentgenogram on a 14 by 17 film.

The Tuberculosis Association carried on this intensive follow-up work leading to examination and the obtaining of 14 by 17 roentgenograms of those individuals requiring review. As a result, 335 persons, or 4.16 per 1,000 examined, were found to have clinically significant tuberculosis. Three-fourths of these 335 were minimal cases, an indication that their chances of recovery are excellent. Presumably the majority of these minimal cases would not have become known at this time unless the mass survey had been held. A check of health department records revealed that 23, or approximately 7 per cent, of the 335 clinically significant cases had been previously reported.

In addition to the 335 cases of tuberculosis which were found, 388 diagnoses of nontuberculous abnormalities were made as a result of the survey. This finding must be considered as of equal importance to the discovery of 335 cases of tuberculosis.

One of the most significant results of this study is the finding that the proportion of cases discovered increases directly with the age of the persons examined.

If one considers all types of pathological cases found among the 80,512 persons examined, it becomes apparent that one per cent of the entire group presents some abnormality requiring medical attention.

A mobile chest roentgenographic service should continue to be a valuable component of any over-all tuberculosis control program. The Erie County, New York, experience demonstrates clearly how such a mobile unit can be coordinated effectively with the official health agencies and other community organizations and facilities.

SUMARIO

Obra de Descubrimiento de Casos de Tuberculosis

Del 4 de marzo de 1946 al 31 de marzo de 1947 se llevó a cabo una obra colectiva de descubrimiento de casos en el Condado Erie, en el Estado de Nueva York, por la Asociación Anti-Tuberculosa de la ciudad de Búffalo y el Condado Erie. Durante dicho período de tiempo, obtuviéronse radiografías torácicas (películas en miniatura) de 80,512 personas, en 1,576 de las cuales se observó la necesidad de estudio más detenido. El primer tiempo en este procedimiento de observación ulterior consistió en un examen y otra radiografía de 35 por 52.5 cm.

La Asociación Anti-Tuberculosa ejecutó esta obra intensa de observación con el examen y la obtención de películas mayores en los individuos que exigían repaso, como resultado de la cual se descubrió que 335 personas, o sea 4.16 por cada 1,000 examinados, mostraban tuberculosis de importancia clínica. Tres cuartas partes de las 335 tenían casos mínimos, indicando esto excelentes probabilidades de curación. Es de presumir que la mayoría de dichos casos mínimos no hubieran sido conocidos para aquella fecha, de no haberse verificado la encuesta colectiva. El estudio de los archivos del departamento de sanidad reveló que 23, aproximadamente 7 por ciento, de los 335 casos clínicamente significativos habían sido denunciados anteriormente.

Además de los 335 casos tuberculosos descubiertos, se hicieron, a consecuencia de la encuesta, 388 diagnósticos de anomalías no tuberculosas, hallazgo este que debe considerarse igual en importancia al descubrimiento de los casos tuberculosos.

Uno de los resultados más importantes de este estudio consiste en la observación de que la proporción de casos descubiertos aumenta en razón directa a la edad de las personas examinadas.

Tomando en cuenta todas las formas de patología observadas entre las 80,512 personas examinadas, resulta evidente que uno por ciento del grupo completo presentaba alguna anomalía que exigía asistencia médica.

Un servicio movible de radiografía torácica debe continuar formando un valioso componente de toda obra general de lucha antituberculosa. Lo observado en el Condado Erie demuestra claramente la forma en que una unidad movible como la descrita puede coordinarse eficazmente con los organismos oficiales de sanidad y con otros organismos y recursos cívicos.

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THE ISOLATION OF THREE DIFFERENT PROTEINS AND TWO POLYSACCHARIDES FROM TUBERCULIN BY ALCOHOL FRACTIONATION. THEIR CHEMICAL AND BIOLOGICAL PROPERTIES[1,2]

FLORENCE B. SEIBERT

INTRODUCTION

Previous studies (1) have shown that the protein moiety of tuberculin is a complex mixture of proteins. The conclusions reached in those studies were based upon researches on fractions isolated from heated tuberculin. Such fractions must have been mixtures of proteins denatured by heat and undenatured proteins that withstood the heat. It was decided, therefore, that a more logical approach to the study of the proteins could be made by fractionating raw unheated tuberculin and later, if desired, denaturing the isolated fractions.

The following outline represents a compilation of the methods which have proved to be most successful in fractionating the tuberculin component into electrophoretically distinguishable substances. No attempt is made to describe in detail each of the many complete fractionations that were made. Complete data are available in files in this laboratory. Only those steps are given which were finally chosen as most desirable after numerous modifications, and it is recognized that the scheme of fractionation here outlined should be considered preliminary. It is presented at this time because it may lead to the development of a more specific tuberculin than has been available previously and because it might be helpful to other investigators in this field.

It will be shown below that the following components of tuberculin were isolated: two distinct polysaccharides, designated I and II; and three distinguishable proteins, designated A, B and C. These proteins have also been discussed previously (1). The A protein had the slowest migration when studied in electrophoresis, and the B and C proteins had a faster migration. While the mobilities of the B and C proteins were approximately the same, their solubility properties were different. Polysaccharide I was the small polysaccharide molecule studied extensively in the past (1, 2), and polysaccharide II was a large molecule not hitherto described and found in quantity only in culture filtrates of certain strains of tubercle bacilli.

OUTLINE OF FRACTIONATION PROCEDURE

Growth of bacilli: Tubercle bacilli of the following strains, human DT, H 37 and A 33 and bovine BCG were grown on Long's synthetic medium, usually for eight to twelve weeks. In the case of one fractionation (No. 99) the bacilli were grown for only five weeks, and in another (No. 93) for thirteen weeks.

[1] From The Henry Phipps Institute, University of Pennsylvania, Philadelphia.

[2] Aided by grants from the Committee on Medical Research of the National Tuberculosis Association.

Harvesting of bacilli: The bacilli were filtered off, without previous heating of the culture, through large Seitz filters as quickly as possible. The filtrates were removed to a cold room held at 1 to 2°C and all subsequent procedures were carried out at this temperature, using no preservative.

Concentration of the filtrate and washing: The filtrates were concentrated by means of ultrafiltration through 9 per cent[3] gun cotton membranes, as described previously (3), to a tenth or a hundredth of the original volume, depending upon the viscosity of the solutions. Toward the end of the concentration water was added and the ultrafiltration continued in order to reduce the salt content of the solutions, as better fractionation occurred when the precipitations were made in the presence of a low salt concentration.

Precipitation of C protein: The C protein was precipitated by adjusting the concentrated ultrafiltrates to pH 3.8 to 4.7 by slowly adding acetic acid with stirring. The precipitates, which were colored a moderate yellow, were centrifuged off and washed with acetate buffer pH 4.0, $\mu = 0.02$. Further purification can be effected by redissolving and reprecipitating.

Fractionation of the supernatant: The supernatants, after removal of the C protein at about pH 4.0, showed in electrophoresis (see figure 4) the presence of two main protein components, designated as A and B proteins. In some cases there was present also a considerable amount of polysaccharide with very low mobility, as can be seen in the last two diagrams of figure 4.

Separation of the A and B proteins was at first attempted by means of ammonium sulfate fractionation, as small amounts of relatively pure A protein had previously been obtained in this way (4, 5). But after repeated trials, using one-fourth, one-third or one-half saturated ammonium sulfate at either pH 4.0 or pH 7.0, it was concluded that this method of separation was not satisfactory.

Therefore, electrophoretic separation was considered. This method is possible only when the resolution of the components is sufficiently sharp to yield distinct components, as in the case of the second supernatant shown in figure 4. The results of fractionation of this supernatant (lot No. 84) will be described later. In the great majority of cases, however, this method is not advisable.

Finally, alcohol fractionation was attempted and proved to be superior to any methods so far used. The steps in the procedure found to be most successful are given below. Although the fractionations were made at a temperature of 1°C, a lower temperature (-5° to -10°C) would be desirable in order to reduce the possibilities of denaturing the proteins.

Precipitation of polysaccharide II: The supernatant, after precipitation of the C protein at pH 4.0, was brought back to neutrality by slow addition of weak alkali while stirring. In cases where the solution was not clear it was filtered through the Seitz filter. It was then concentrated on fresh ultrafilters to a small volume. To this solution was added sufficient 95 per cent alcohol with stirring to give a 30 per cent alcohol concentration. A white gelatinous precipitate occurred which dissolved slowly in water, giving a white opalescent solution which appeared blue in dim light. It was named polysaccharide II to distinguish it from the polysaccharide previously described (1, 2) and now called polysaccharide I.

Precipitation of protein B: The supernatant from polysaccharide II, which had a pH of 7.0 and an alcohol concentration of 30 per cent, was readjusted to about pH 4.6 by

[3] Nine per cent gun cotton, as now available, gives a porosity which was given previously by 11 per cent.

addition of acetic acid with stirring and a precipitate, consisting chiefly of protein B, came out.

Precipitation of protein A: After removal of the B protein at about pH 4.6 with 30 per cent alcohol, more alcohol was added to the supernatant to give a concentration of 70 per cent. The resulting precipitate contained chiefly the A protein.

Recovery of polysaccharide I from final residue: After the A protein was removed, the supernatant was dialyzed to remove alcohol and then concentrated by ultrafiltration. Any trace of protein was precipitated by trichloracetic acid and removed. The supernatant was again dialyzed to free it from trichloracetic acid and the remaining solution contained polysaccharide I.

TABLE 1

Yields of proteins and polysaccharides isolated from different preparations of tuberculin

LOT NUMBER OF PREPARATION	STRAIN* OF TUBERCLE BACILLUS	VOLUME OF LONG'S MEDIUM INOCULATED	POLYSACCHARIDE I	POLYSACCHARIDE II	PROTEIN A	PROTEIN B	PROTEINS A + B	PROTEIN C
		Liters	*Gm.*	*Gm.*	*Gm.*	*Gm.*	*Gm.*	*Gm.*
84	DT	14	0.6				0.7	0.4
88	DT	20	0.4				0.6	0.7
90	DT	20	2.8				1.1	2.5
92	H37	19	0.7?	4.7			1.5	2.0
93	H37	4.5		3.6			1.2	0.8
94	DT	10	0.9				0.6	3.4
95	BCG	40	0.4	0.2			2.4	1.1
96A	A33	24.5	1.0	0.7	1.7	0.32		6.3
96B	A33	9.4	0.2+	0.1			0.4	1.5
98	DT	36	0.5	0.1	0.9	2.7		4.7
99	DT	35			0.025	0.03		0.2

* All strains were human type tubercle bacilli except BCG.

EXPERIMENTS

Yields of Components Isolated

Table 1 shows the amounts of the different proteins and polysaccharides actually isolated during each fractionation. No claim is made that the maximum yield of any of the products was obtained. During these experiments the proper conditions for isolation were being sought and the losses were necessarily great. This is especially true in the case of polysaccharide I. The yields do, however, give a rough idea of the relative amounts of the different components present and they show that in most cases the C protein constituted a large portion of the total yield.

Electrophoretic Analyses of the Isolated Fractions and Components

All electrophoretic analyses were carried out in phosphate buffer pH 7.7, $\mu = 0.1$, at a potential gradient of 9 to 10 volts per cm. The ranges of the mobilities[1] of the isolated components are shown in table 2. When purer fractions

[1] All mobilities are recorded in units of 10^{-5} cm.^{2} volt^{-1} sec.$^{-1}$.

are obtained these ranges undoubtedly will become narrower. Characteristic properties important in the isolation of these components are also given in table 2.

Concentrated original ultrafiltrates: Examination of small samples of the original filtrates by means of electrophoresis afforded a quick method for revealing the relative proportions of the different components present (figure 1). It can be seen, for example, that the filtrates from lots Nos. 84, 98 and 99, all made from the human strain DT, contained relatively more of the faster migrating components, the proteins, than did the filtrates from lots Nos. 92 and 96A, made respectively from the H 37 and A 33 human strains. The latter two filtrates, on the other hand, contained a very large proportion of the slowly migrating components, which have proved to be the polysaccharides.

New components are also readily revealed by means of these electrophoresis analyses. For example, the diagram of the filtrate of lot No. 99 (figure 1)

TABLE 2

Electrophoretic mobilities, solubilities and conditions of precipitation of the isolated proteins and polysaccharides

COMPONENT	*MOBILITY × 10^{-4} CM², VOLT⁻¹ SEC⁻¹	SOLUBILITY AT pH 3.8–4.7	CONDITIONS FOR PRECIPITATION
Protein			
A	−3.4 to −3.8	soluble	70 per cent alcohol, pH 4.6
B	−5.4 to −6.4	soluble	30 per cent alcohol, pH 4.6
C	−6.1 to −7.3	insoluble	acetic acid, pH 4.0
D	−8.6	insoluble	acetic acid, pH 4.0
Polysaccharide			
I	−1.4 to −2.0	soluble	95 to 100 per cent alcohol
II	−1.4 to −1.6	soluble	30 per cent alcohol, pH 7.0

* Mobilities in phosphate buffer, pH 7.7, $\mu = 0.1$, and potential gradient of 9 to 10 volts per cm.

showed a significant amount of a component which migrated with a greater velocity than the proteins. As the only difference between this filtrate and several other filtrates from the same (DT) strain, as well as from other strains, was that the culture was only five weeks old, in contrast to the eight to ten weeks of usual growth, it is possible that this component may be characteristic of early growth. Its nature will be discussed below under protein C.

Protein C: This protein was always readily precipitated at a pH 3.8 to 4.7 and even after a single precipitation showed one main electrophoretic component with a mobility of −6.1 to −7.3. Figure 2 shows diagrams of six such protein preparations isolated from the culture filtrates of four different strains of tubercle bacilli.

The fraction which precipitated at pH 4.5 from the culture filtrate (lot No. 99) of only a five weeks old culture of tubercle bacilli, strain DT, contained, in addition to C protein with mobility of −6.4, another component with a greater mobility, −8.6 (figure 3). This mobility is characteristic of nucleoproteins and

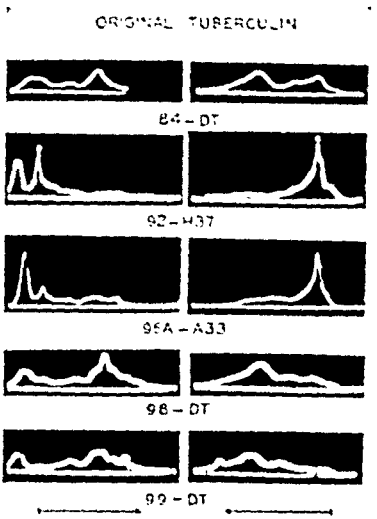

FIG. 1. Electrophoretic diagrams of six samples of concentrated original tuberculins. The diagrams on the right represent the descending patterns and those on the left the ascending patterns. After about two hours electrophoresis the components are migrating in the directions indicated by the arrows, those with the fastest mobilities occupying the positions toward the heads of the arrows. The anomalous conductivity boundaries, δ and ε, which are the slowest boundaries seen on either side toward the end of the arrows, are larger on the ascending side (δ) and relatively inconspicuous on the descending side (ε). Often the polysaccharides separate very poorly or not at all from these boundaries.

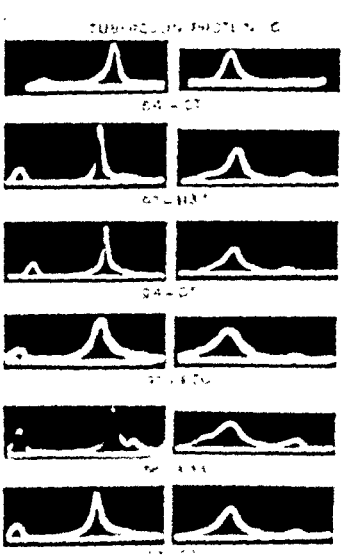

FIG. 2. Electrophoretic diagrams of six isolated C proteins which appear as the major components on both ascending and descending patterns.

it may be significant that this component appeared only in considerable quantity in the young cultures. It has been designated D component.

Supernatant from C protein, containing A and B proteins: After the C protein was removed at about pH 4.0, there remained in solution the polysaccharides and some soluble proteins, which when examined by electrophoresis appeared to consist of two proteins. One of them had nearly the same mobility as the C protein and was called B protein. The other one migrated more slowly and was called

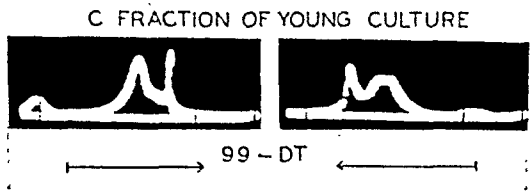

FIG. 3. Electrophoretic diagrams of the C fraction isolated from a young culture, showing the extra D component as the faster component above the head of the arrows.

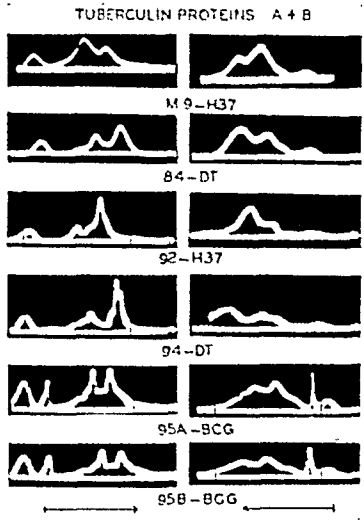

FIG. 4. Electrophoretic diagrams of six supernatant fractions after removal of the C protein. In each case the two proteins, B and A, are seen as the two fastest components. In the last two diagrams an additional slow component appears as a sharp peak with slowest mobility, next to the anomalous δ and ϵ boundaries, with no mobility. These are the polysaccharide II.

A protein (figure 4). These same proteins were found six different times and in filtrates from three different strains of tubercle bacilli.

Protein B. This protein was isolated in two different ways, as described above under the fractionation procedure.

Separation by electrophoresis. When the resolution in electrophoresis of the A and B proteins was good, as in the case of the supernatant from lot No. 84 (figure 4), the B protein was isolated by removing mechanically the faster migrating component in the ascending limb of the electrophoresis cell. It was sent to Mrs. Ellen Bevilacqua at the University of Wisconsin for sedimentation and diffusion determinations. The results were reported in her thesis (6) and showed

that it was homogeneous in both sedimentation[5] and diffusion, with the following constants, S = 2.0, D = 9.6, and a/b ratio of 5.0, and a molecular weight of about 20,000. The sedimentation constant corresponds with that found for the faster migrating protein separated by electrophoresis in an earlier study (4).

Separation by alcohol precipitation. After precipitation of the C protein at 4.0, polysaccharide II was precipitated by 30 per cent alcohol from the supernatant which had been adjusted to a neutral reaction (see above under procedure). When this polysaccharide II was also removed, the clear supernatant,

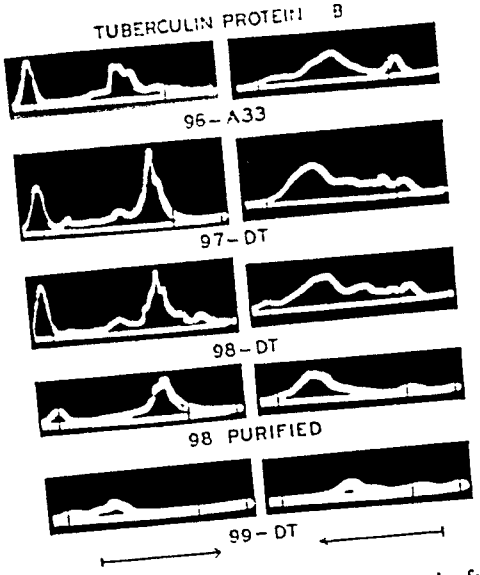

FIG. 5. Electrophoretic diagrams of five isolated B protein fractions. The B proteins appear as the conspicuous faster components. The smaller slower components are A protein and polysaccharides, and the occasional small fastest one is the D component.

containing 30 per cent alcohol, was again readjusted back to about pH 4.6 and a fraction containing mostly protein B precipitated. Fractions with similar mobilities were isolated four different times and from two different strains of tubercle bacilli, as may be seen in figure 5. In all cases the largest component represents B protein and in most cases the diagrams show also the presence of a smaller amount of a component or components migrating at a slower rate than the B protein. These components are probably the A protein and polysaccharides. Therefore, in one case (lot No. 98) where there was sufficient material available, further purification was undertaken. The entire procedure for isolation was repeated and the resulting product did show the presence of only very little of the contaminating components (figure 5, diagram marked "98 purified").

[5] Sedimentation constants (S) are recorded in units $\times 10^{-13}$, and diffusion constants (D) in units of $\times 10^{-7}$.

Protein A: This protein also has been isolated in two different ways, viz., by (1) electrophoresis, and (2) alcohol fractionation.

Separation by electrophoresis. When the supernatant remaining after the removal of the C protein was subjected to electrophoresis, the fraction migrating with the slowest mobility remained behind on the descending side and could easily be withdrawn. This fraction contained the protein with the slowest mobility, which is called protein A, and also the slowly migrating polysaccharides. When studied by Mrs. Bevilacqua (6) in the ultracentrifuge it also proved to be heterogeneous and gave evidence of at least two components, for one of which a sedimentation constant of S = 1.8 was calculated.

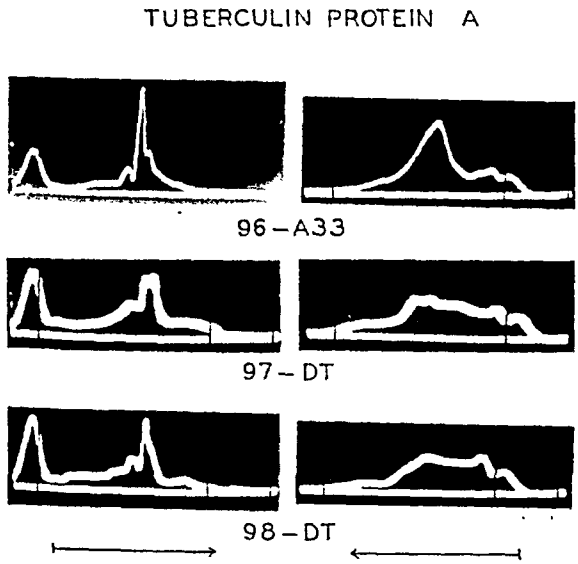

FIG. 6. Electrophoretic diagrams of three isolated A protein fractions. The conspicuous components represent the A proteins. The descending diagrams show also the presence of a considerable amount of slower components, the polysaccharides.

Separation by alcohol fractionation. After removal of the B protein fraction (see above) the supernatant had a pH of about 4.6 and an alcohol concentration of 30 per cent. The alcohol concentration was then increased to 70 per cent, as described under procedure, and a fraction containing most of the A protein precipitated (figure 6). In this precipitate only a trace of the faster migrating protein, B, was present, but there were considerable amounts of the very slowly migrating polysaccharides. Further purification of this fraction is, therefore, desirable but has so far not been possible because of lack of material.

This fraction when dissolved in water gave clear colorless solutions. It is possible that the A protein is the protein with molecular weight about 35,000 to 42,000 (4, 5) and it may also be the one which was crystallized (7), but further data will be necessary to establish such identity.

Polysaccharide II: This polysaccharide has not hitherto been described and appeared in considerable quantity only in the culture filtrates of certain strains

of tubercle bacilli, *viz.*, the H 37, A 33 and BCG strains. Its chemical and biological properties will be described in another publication. When present it could be distinguished by the opalescence which it gave to the culture filtrates. It was

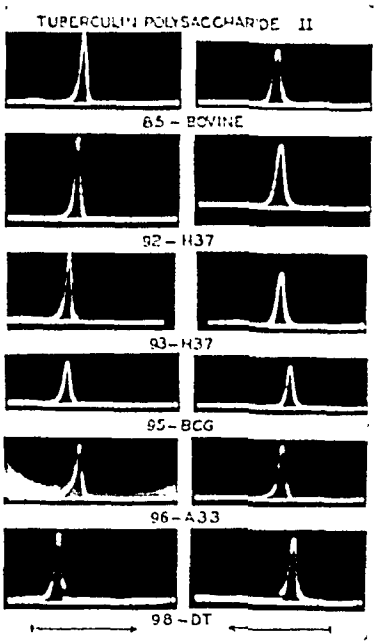

FIG. 7. Electrophoretic diagrams of six isolated polysaccharide II preparations taken after four hours of electrophoresis. No other component appeared.

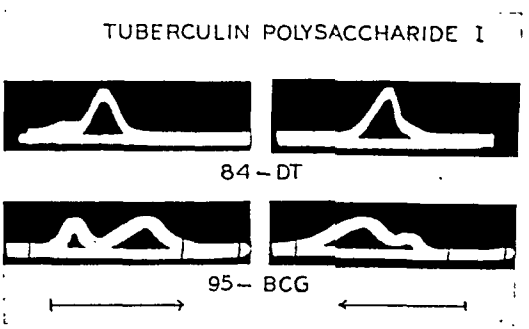

FIG. 8. Electrophoretic diagrams of two isolated polysaccharide I preparations. The δ and ε boundaries are the slower inconspicuous boundaries in the first pattern and the more definite slower peaks in the second pattern.

readily precipitated by 30 per cent alcohol at neutrality from the supernatant of the C protein, as described above. When studied in electrophoresis (figure 7) it appeared to be homogeneous, even after being subjected to the current for four hours. It has been isolated six times from the filtrates of five different strains of tubercle bacilli and its presence was apparent in the original concentrated culture

filtrates as the sharp peak with very low mobility, which can be seen in the electrophoretic diagrams of lot No. 92 H 37 and lot No. 96 A–A 33 in figure 1.

Polysaccharide I: After all of the above proteins and polysaccharide II were removed from the culture filtrate, the residue contained a considerable amount of this polysaccharide (1). It could be isolated as described above under fractionation procedure. It was the highly soluble polysaccharide of low molecular weight (7,000 to 9,000) which had been studied extensively and whose properties have been reviewed previously (1, 2). The electrophoretic diagrams are shown in figure 8. In most cases it would be possible to isolate considerably more of this component than is reported in table 1.

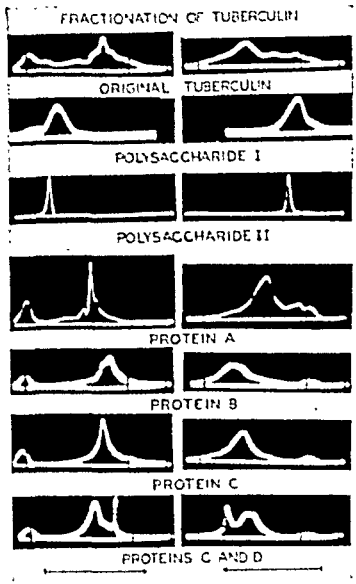

FIG. 9. Electrophoretic diagrams of a typical original concentrated tuberculin and below it the isolated fractions corresponding to those distinguishable in the original diagram. As all pictures were taken after approximately the same time of electrophoresis, their positions indicate their relative rates of migration. The sizes of the components do not correspond with the relative concentrations present in the original tuberculin.

Figure 9 shows an electrophoretic diagram of an original concentrated tuberculin and below it patterns of isolated components with mobilities corresponding to those distinguishable in the original composite diagram.

Chemical Analyses

Methods. The content of polysaccharide was determined by means of the carbazole reaction (8, 9), using the purest polysaccharide I or polysaccharide II as standard.

Nucleic acid was determined by means of the Dische diphenylamine reaction (8, 10), using Hammarsten's sodium thymus nucleate as standard.

Both carbohydrate and nucleic acid contents were calculated in relation to a simultaneous concentration of 1 mg. per ml. of protein in the same solutions.

The spectral absorption in ultraviolet light was measured with a Beckman spectro-

photometer—U V, and the specific densities were calculated on the basis of a protein concentration of 1 mg. per ml. The protein contents were determined from the nitrogen contents.

TABLE 3

Polysaccharide and nucleic acid analyses and spectral densities of tuberculin proteins, A, B and C.

TYPE OF PROTEIN	LOT NUMBER	POLYSACCHARIDE	NUCLEIC ACID	SPECIFIC SPECTRAL DENSITY AT λ 2800 Å	SPECIFIC SPECTRAL DENSITY AT λ 2500 Å	RATIO Dλ2800/λ2500	TUBERCULIN SKIN POTENCY RELATIVE TO PPD-S IN HUMAN BEINGS
		mg. per ml.	*mg. per ml.*				
A + B	90			1.18	0.39	3.1	>
A + B	92			2.01	0.75	2.7	
A + B	93			1.07	0.54	2.0	
A + B	94			1.36	0.56	2.5	>
A + B	95	0.154	0.0026	1.01	0.48	2.1	>
A	96A*		0.0017	2.07	0.79	2.6	<
A	96B*		0.0100	1.29	0.59	2.2	>x
A	97		0	1.52	0.48	3.2	>x
A	98	1.420	0.0106	1.34	0.58	2.3	=
A + B	99‡		0.0090	0.74	0.41	1.8	>x
Average				1.36	0.56	2.5	
B	96A*		0.0053	0.88	0.47	1.9	<x
B	97		0.0060	1.42	0.70	2.0	>x
B	98			1.39	0.70	2.0	>x
Average				1.85	0.94	2.0	
C	90			2.14	1.54	1.4	
C	92			1.34	0.87	1.5	<
C	93			1.58	1.11	1.4	
C	94			1.79	1.41	1.3	<
C	95	0.070	0.0067	1.69	1.21	1.4	<
C	96A*		0.0545	2.08	1.80	1.2	<
C	96B*		0.0040	1.37	0.90	1.5	>x
C	98	0.034	0.0035	1.92	1.50	1.3	<
C + D	99‡		0.0070	0.99	0.51	1.9	<x
Average				1.64	1.20	1.4	

* Lots 96 A and B were made from thirteen week old cultures and the A protein was precipitated with trichloracetic acid instead of acetic acid.

‡ These fractions were made from a five weeks old culture filtrate. See text.

x These tests were made in tuberculous guinea pigs rather than in human beings.

Results: The concentrations of nucleic acid and of carbohydrate are recorded in table 3. The few analyses available show a higher content of polysaccharide with the A and A + B proteins than with the C proteins, as would be expected. The content of nucleic acid was very low in all cases, but as there may be an error

when the diphenylamine reaction is used in the presence of much protein, further work must be done before a definite statement can be made concerning the exact content of nucleic acid in these fractions.

The specific spectral maximum and minimum densities in the ultraviolet light, as well as the ratios of the maximum to the minimum densities, are recorded in table 3. The maximum absorption was at a wave length of 2,800 Å, except in a few cases of the C protein, where it was at a wave length of 2,700 Å, and minima were in all cases at a wave length of 2,500 Å. It is apparent from table 3 that, whereas the maximum specific densities at a wave length of 2,800 Å were nearly the same for all A or C proteins, there was considerable difference between the A and C proteins in the amount of specific density at their minima, *i.e.*, at a wave

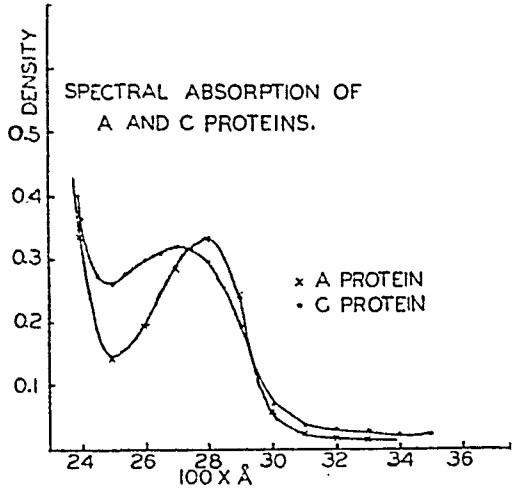

FIG. 10. Spectral absorption curves of the A and C proteins, measured with a Beckman spectrophotometer in the ultraviolet region. The concentrations chosen were such as to give practically equivalent densities at a wave length of 2800 Å, in order to show the relative difference in densities at 2500 Å.

length of 2,500 Å. This is illustrated by one set of curves given in figure 10. This difference caused a large difference in the ratios of density at 2,800 Å/2,500 Å for the A and C proteins, and means that there is something in the C proteins which has about twice as great an absorption capacity at a wave length of 2,500 Å as in the A proteins. The presence in the C proteins of a substance such as nucleic acid, with its maximum absorption at a wave length of 2,600 Å could be responsible for this effect, as well as for causing the maximum to shift from a wave length of 2,800 Å to 2,700 Å. Such a nucleoprotein would also have a higher electrophoretic mobility, similar to that found for the C proteins.

Tuberculin Potency of the Proteins and Polysaccharides

The tuberculin potencies of the isolated proteins, given in equivalent dosages in 0.1 ml. based on the nitrogen contents, were compared in tuberculous guinea pigs and in tuberculin-positive human beings. The size of skin reaction elicited by the proteins was compared with that caused by an equal dosage, based on the

nitrogen content, of the PPD-S, which is the tuberculin adopted as standard in the United States (3). In some cases only one product was compared on the same guinea pig with the standard, and in other cases three or more proteins were simultaneously injected with the standard. The results are given in table 4. All reactions were read twenty-four hours following the tests and these reactions were recorded in the top lines in the table. Readings were also made at forty-eight hours and those reactions recorded in the lower lines.

The results of the multiple test procedure showed that the A proteins always caused larger reactions than did the C proteins, while usually the B proteins gave reactions of intermediate size. When the A and C proteins alone were compared with each other, the A was again the more potent.

When tested in sensitive human beings in a dosage of 0.00002 mg., the A + B proteins were more potent than the PPD-S, and the C proteins less potent. However, when tested in less sensitive individuals in a dosage of 0.005 mg., the A + B proteins were more potent than the PPD-S and the reactions to the C proteins were as great or greater than with the PPD-S, in spite of the fact that some reactions were even missed with it. These reactions have been reported in another paper (11). This was interpreted to mean that the C protein was less specific and the A protein more specific than the whole mixture of proteins existing in PPD-S.

Neither of the polysaccharides gave skin reactions in tuberculous animals or human beings. This was the conclusion reached by Watson (2, 12) with regard to polysaccharide I. In the present study it was found that polysaccharide II also elicited no reaction in tuberculous guinea pigs nor (even with 0.05 mg. dose) in sensitive human beings who gave definite tuberculin reactions with 0.00002 mg. PPD-S.

DISCUSSION

The indication that the A protein might give more specific tuberculin reactions than the other proteins in tuberculin is sufficient reason for further endeavor to obtain this protein in highly purified form. It has been shown in these studies that the A type of protein is the most potent in causing the tuberculin skin reaction and this confirmed the earlier finding (4) that that protein fraction in crude tuberculin which migrated with the slowest mobility was the most potent one in both the skin test and the lethal test in tuberculous guinea pigs.

In view of the statement by Lind (13) that a more potent PPD could be made from young than from old cultures, it was hoped that it might be possible to isolate relatively more of the A type of protein from such cultures. This was not found to be the case, however, as shown in the yield of A protein isolated from the filtrate of lot No. 99 (see table 1), nor were any of the isolated fractions relatively more potent than similar protein fractions obtained from older cultures. As a matter of fact, all components were isolated from this five-week-old culture filtrate in relatively the same proportion as from older cultures of the same strain (DT), but in insignificant amounts. The only difference found was the notable presence of the component (D) which migrated with a high velocity, suggestive

TABLE 4

Comparison of skin reactions in guinea pigs with different tuberculin proteins

CONDITION OF GUINEA PIGS	NUMBER OF GUINEA PIGS TESTED	DOSAGE	AVERAGE DIMENSIONS OF REACTIONS IN MILLIMETERS WITH				
			PPD-S	A (Lot 98)	B (Lot 98)	B (Purified) (Lot 98)	C (Lot 98)
		mg.					
Normal	2	0.005	0 0	0 0	0 0	0 0	0 0
BCG	3	0.005	18 x 22 x 2.3 19 x 22 x 3.0	27 x 38 x 3.3 21 x 31 x 3.0	25 x 30 x 2.6 22 x 22 x 3.0	26 x 29 x 3.0 21 x 25 x 3.0	20 x 20 x 2.6 20 x 21 x 3.0
Normal	2	0.0005	0 0	0 0	0 0	0 0	0 0
Tuberculosis	3	0.0005	18 x 21 x 2.7 16 x 16 x 2.7	26 x 32 x 3.0 23 x 24 x 3.7			
Tuberculosis	4	0.0005	25 x 31 x 3.0 21 x 19 x 2.7			29 x 35 x 3.7 23 x 25 x 3.3	
Tuberculosis	3	0.0005	18 x 23 x 2.7 18 x 18 x 2.7				22 x 23 x 3.0 20 x 22 x 3.3
Tuberculosis	2	0.0005	14 x 17 x 2.0 17 x 19 x 2.6	20 x 22 x 3.0 23 x 24 x 3.7		18 x 20 x 2.6 18 x 19 x 3.0	11 x 13 x 1.3 13 x 13 x 2.0
Tuberculosis	3	0.0005		21 x 22 x 3.0 22 x 23 x 3.7			15 x 15 x 2.3 15 x 17 x 2.3
			PPD-S	A (Lot 99)	B (Lot 99)		C (Lot 99)
Normal	2	0.0005	0 0	0 0	0 0		0 0
Tuberculosis	3	0.0005	14 x 18 x 2.3 14 x 16 x 2.0	21 x 24 x 3 19 x 24 x 3			
Tuberculosis	3	0.0005	18 x 20 x 3.0 19 x 19 x 3.0		17 x 21 x 3.0 15 x 18 x 3.0		
Tuberculosis	3	0.0005	21 x 24 x 3.0 17 x 17 x 3.0				22 x 26 x 3.0 15 x 18 x 2.7
Tuberculosis	2	0.0005	21 x 23 x 3.0 20 x 18 x 3.0	22 x 25 x 3.0 21 x 22 x 3.0	20 x 21 x 3.0 19 x 19 x 3.0		18 x 19 x 2.5 17 x 18 x 2.0
			PPD-S	A (Lot 97)	B (Lot 97)		
Normals	2	0.00025	0 0	0 0	0 0		
Tuberculosis	4	0.00025	17 x 17 x 2.5 16 x 17 x 2.5	19 x 20 x 3.3 18 x 20 x 2.8	22 x 23 x 3.3 20 x 22 x 3.3		
			PPD-S	A (Lot 96A)	B (Lot 96A)		C (Lot 96A)
Normals	2	0.00025	0 0	0 0	0 0		0 0
Tuberculosis	4	0.00025	19 x 20 x 3.0 17 x 17 x 3.0	21 x 23 x 3.2 19 x 21 x 3.0	18 x 19 x 3.0 15 x 16 x 2.5		17 x 18 x 2.7 15 x 17 x 2.5
			PPD-S	A (Lot 96B)			C (Lot 96B)
Normals	2	0.00025	0 0	0 0			0 0
Tuberculosis	4	0.00025	16 x 18 x 2.3 14 x 15 x 2.3	20 x 22 x 3.3 19 x 21 x 3.0			19 x 21 x 2.5 14 x 16 x 2.3

of nucleoprotein. The C protein fraction which contained this component (figure 2) was also not significantly more potent than other C proteins. It is interesting in this connection that Heilman (14) noted also that the protein associated with nucleic acid was relatively less potent as a specific cytotoxic agent.

SUMMARY

A preliminary procedure, utilizing fractionation with alcohol at low temperature, has been presented for isolating from raw unheated tuberculin, made by growing different strains of tubercle bacilli on synthetic medium, two different polysaccharides (designated I and II) and three different proteins (designated A, B, and C).

Each of these components, all of which have been isolated several times and from the culture filtrates of different strains of tubercle bacilli, can be identified by its electrophoretic mobility. In the order of increasing mobility they were polysaccharides I and II, proteins A, B, and C.

A component with a still greater mobility, designated as D and possibly a nucleoprotein, was found to exist in significant amount in a five-week-old culture but not in older cultures from the same strain or other strains of tubercle bacilli.

Different strains of tubercle bacilli yielded different relative amounts of the polysaccharide and protein components. The amounts could be predicted from the electrophoretic diagrams of the original concentrated tuberculin filtrates.

Protein A was more potent and possibly more specific than protein C in eliciting the tuberculin skin reaction in infected animals and human beings.

The A and C proteins had similar spectral specific densities in ultraviolet light at a wave length of 2,800 Å, but the C proteins had nearly twice the specific densities at a wave length of 2,500 Å, giving a density ratio $\left(\frac{\lambda\ 2800}{\lambda\ 2500}\right)$ of about half that for the A proteins. Utilization of this ratio may help in the identification of A and C proteins during a fractionation.

SUMARIO

Aislamiento por la Fraccionacion con Alcohol de Tres Distintas Proteínas y Dos Polisacáridos de la Tuberculina, con las Propiedades Quimicas y Biologicas de los Mismos

Descríbese un procedimiento preliminar, que utiliza la fraccionación con alcohol a baja temperatura, para aislar de la tuberculina bruta no calentada (obtenida cultivando en medio sintético diversas cepas de bacilos tuberculosos) dos distintos polisacáridos (designados I y II) y tres diversas proteínas (denominadas A, B y C).

Cada uno de dichos componentes, todos los cuales han sido aislados varias veces y de los filtrados culturales de diversas cepas tuberculosas, puede ser identificado por su movilidad electroforética, figurando en el orden de su mayor movilidad los polisacáridos I y II y las proteínas A, B y C.

Otro componente, dotado aun de mayor movilidad, y denominado D, posible-

mente una núcleoproteína, se encontró en cantidad significativa en un cultivo de cinco semanas, pero no en los cultivos más viejos de la misma cepa ni en otras cepas de bacilos tuberculosos.

Acknowledgment

The writer wishes to express her appreciation to Dr. J. W. Williams for discussions concerning methods of alcohol fractionation and to Mrs. Ellen Bevilacqua for the sedimentation and diffusion determinations made in Doctor Williams's laboratory.

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A COMPARISON OF THE IMMUNOGENICITY FOR GUINEA PIGS OF BCG CULTURED INTERMITTENTLY AND CONTINUOUSLY IN THE PRESENCE OF BILE[1]

IRWIN S. NEIMAN AND NELDA HOLMGREN

INTRODUCTION

In their well-known textbook, Topley and Wilson (1), after reviewing the technique of BCG cultivation, point out that the virulence of BCG is continually falling and that the possibility exists that under such circumstances its protective value might disappear in time. It is important to recall in this connection the method of cultivation of BCG prescribed by Calmette (2). Briefly, it was recommended that BCG be cultured on a glycerinated potato slab in Roux tubes, the bottom portion of which contained 5 per cent glycerin at pH 7.0 to 7.2. At regular intervals the culture was returned to a similar tube containing a potato slab treated with ox-bile glycerin mixture and, in the bottom portion, 5 per cent glycerin in ox-bile. Calmette believed that the reduction in the virulence of BCG resulted from cultivation in the presence of ox-bile. Griffith (3) was able to show that a pathogenic bovine strain of the tubercle bacillus did lose virulence under such conditions of cultivation.

Holm (4), in discussing the use of BCG in Denmark, makes the statement that the culture kept at the State Serum Institute in Copenhagen is transferred only occasionally to a medium containing ox-bile and only then when it is apparent that BCG is "weakened in virulence." Moreover, he states ". . . a new increase in virulence was obtained by several passages on bile potato." It is difficult to understand this assertion in the light of Calmette's original thesis and of Griffith's (3) work. One would expect that the return to cultivation on bile-potato medium would act in a reverse fashion.

In the BCG laboratory of the Municipal Tuberculosis Sanitarium of Chicago, in addition to the culture used for routine vaccination purposes, a strain of the organism has been maintained continually on ox-bile-glycerin-potato medium. It has been conscientiously transferred at two week intervals from 1934 to the present. In order to determine whether this treatment resulted in a loss in immunogenic ability, an experiment was performed which is detailed below.

MATERIALS AND METHODS

The culture of BCG used in these experiments has been kept under rigorous control since its arrival in this country in 1934. It has been cultivated by the methods prescribed by Calmette (2) and transferred at two week intervals. Thousands of guinea pigs have been used to test its avirulence and at no time has any evidence been apparent that this strain can cause progressive tuberculosis in guinea pigs. Intraperitoneal inoculation of several milligrams of the organism into guinea pigs results in, at most, the appearance of tubercles in the omentum and occasional tubercles in the liver and spleen. These lesions are self-limiting and usually have disappeared within one year.

Two groups of tuberculin negative (1:10 Old Tuberculin), commercially obtained guinea

[1] From the Department of Microbiology and Public Health, Chicago Medical School and the Municipal Tuberculosis Sanitarium of Chicago.

pigs were vaccinated by the multiple puncture method. On each guinea pig two areas on the back immediately lateral to the midline were shaved and cleansed. On each area a large drop of suspension of the culture was placed and through it 45 separate and discrete punctures were made. One group, composed of 16 animals, was vaccinated with what is designated for the purpose of this report "regular" vaccine, *i.e.*, a suspension of BCG prepared in the usual way for vaccinating human beings. The other group, composed of 18 animals, was inoculated with a suspension prepared from the culture which has been

TABLE 1

Tuberculin reactions of guinea pigs vaccinated with BCG

"REGULAR" VACCINE				"BILE" VACCINE			
Guinea Pig Number	Date Vaccinated	Date tested	Result[1] in mm.	Result[1] in mm.	Date tested	Date vaccinated	Guinea Pig Numbers
2420	11–18–46	1–8–47	$8 \times 8^{tr}_{+}$	$6 \times 6^{+}_{+}$	1–8–47	11–18–46	2402
3449	"	"	$10 \times 10^{+}_{+}$	$8 \times 10^{tr}_{+}$	"	"	2428
2426	"	"	$12 \times 10^{tr}_{+}$	$10 \times 10^{+}_{+}$	"	"	3047
2437	"	"	$12 \times 10^{tr}_{+}$	$8 \times 12^{tr}_{+}$	"	"	2416
3353	"	"	$12 \times 12^{tr}_{+}$	$12 \times 12^{tr}_{+}$	"	"	3027
2494	"	"	$12 \times 12^{+}_{++}$	$12 \times 10^{+}_{+}$	"	"	3028
3281	"	"	$12 \times 12^{+}_{++}$	$12 \times 14^{tr}_{+}$	"	"	3041
2470	"	"	$12 \times 14^{+}_{++}$	$12 \times 14^{+}_{+}$	"	"	2491
2474	"	"	$10 \times 12^{++}_{++}$	$14 \times 12^{++}_{+}$	"	"	3040
2492	"	"	$14 \times 14^{+}_{++}$	$12 \times 10^{++}_{++}$	"	"	2408
3433	"	"	$14 \times 12^{++}_{++}$	$12 \times 12^{++}_{++}$	"	"	3039
3026	"	"	$14 \times 14^{++}_{++}$	$12 \times 12^{++}_{++}$	"	"	2455
3460	"	"	$14 \times 14^{+++}_{+++}$	$14 \times 14^{+}_{++}$	"	"	2403
2443	"	"	$16 \times 14^{++}_{++}$	$18 \times 14^{+}_{+}$	"	"	2440
3051	"	"	$14 \times 16^{+++}_{++}$	$16 \times 12^{++}_{+}$	"	"	3260
3457	"	"	$18 \times 14^{+++}_{+++}$	$16 \times 14^{++}_{++}$	"	"	2454
				$20 \times 14^{++}_{+++}$	"	"	2430
				$16 \times 20^{+++}_{+++}$	"	"	2446

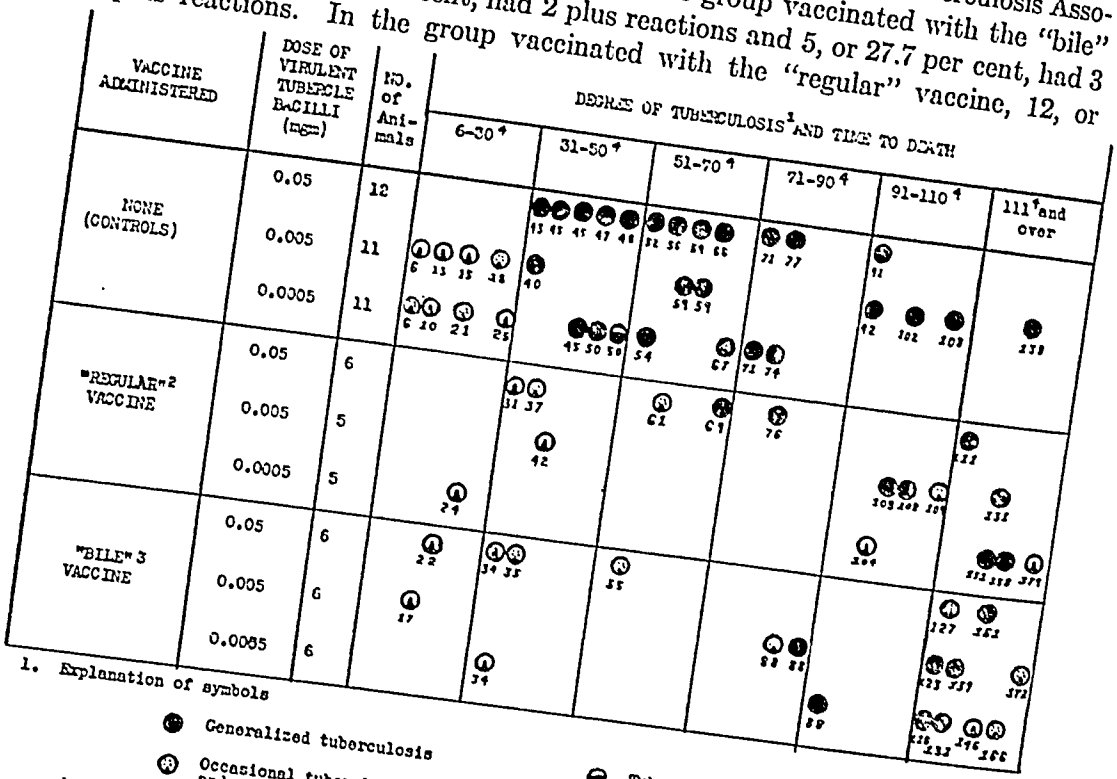
[1] The results of the test were read at forty-eight hours and measured in 2 diameters in millimeters. The degree of redness is indicated by the upper + sign and degree of edema by the lower. The latter takes into consideration the thickness of the edema. Tr. = trace. The reactions are arranged as closely as possible in increasing intensity by the National Tuberculosis Association standards. The 1+ reactions are on the first line, the 2+ reactions on the next twelve lines and the 3+ reactions on the last five lines.

kept continuously transferred in the ox-bile-glycerin-potato medium (the "bile" vaccine). Each vaccine was in a concentration of 15 mg. per cc.

Seven weeks after immunization, each of the guinea pigs was tested for tuberculin sensitivity by an intradermal injection of a 1:10 dilution of Old Tuberculin. One week later all the animals were injected subcutaneously with a suspension of a virulent tubercle bacillus (H_{37}). At the same time 34 normal tuberculin-negative guinea pigs, as controls, were injected with the same suspension and the same dosages of virulent tubercle bacilli. The viability of both the vaccine and H_{37} was controlled by inoculation of Löwenstein's medium at time of use. In each case viability was confirmed.

RESULTS

Following inoculation with BCG all the guinea pigs became positive to tuberculin. The results of the individual tests are noted in table 1. The reactions varied from 1 plus to 3 plus by the standards of the National Tuberculosis Association,[2] and examination reveals that, in the group vaccinated with the "bile" vaccine, 12, or 66.6 per cent, had 2 plus reactions and 5, or 27.7 per cent, had 3 plus reactions. In the group vaccinated with the "regular" vaccine, 12, or



1. Explanation of symbols

Generalized tuberculosis

Occasional tubercles in liver and spleen

Tuberculosis of liver and spleen

Local tuberculosis at point of inoculation only

Numbers under symbols indicate exact time to death in days

2. Vaccine prepared from culture regularly returned to ox-bile-glycerine potato medium after 5 transfers on plain glycerine potato.
3. Vaccine prepared from culture continously kept on ox-bile-glycerine potato medium.
4. Time to death in days after inoculation with virulent tubercle bacilli.

FIG. 1. The protective value of BCG in guinea pigs

75.0 per cent, had 2 plus reactions and 3 or 18.7 per cent had 3 plus reactions. This would seem to indicate that the degree of sensitivity induced by the "regular" vaccine was essentially the same as that induced by the "bile" vaccine. Under the conditions of this experiment, utilizing only a single test dose (10 mg. Old Tuberculin), it is impossible to affirm or deny that a difference in hypersensitivity exists between the two groups, as quantitative tuberculin testing was not done. In this experiment the tuberculin reaction was utilized only to prove the existence of infection engendered by the presence of BCG. For immunization purposes, a positive reaction to tuberculin, irrespective of degree, indicated a "take."

[2] + = an area of edema greater than 5 mm. but less than 10 mm.
++ = an area of edema greater than 10 mm. but less than 15 mm.
+++ = an area of edema greater than 15 mm.
++++ = necrosis irrespective of size.

In order to determine the degree of protection, the animals were divided into three secondary groups. Each group was given a different dose of the challenge tubercle bacillus (H_{37}), *i.e.*, 0.05 mg., 0.005 mg. and 0.0005 mg. In figure 1 are depicted the degree of tuberculosis and time to death of each of the various groups of guinea pigs. It is interesting to note that the three graded doses made remarkably little difference in the average time to death of the controls. Detailed examination of the figure makes it obvious that BCG, whether cultured intermittently or continuously on bile, confers a measurable degree of protection. This is further emphasized by the fact that 12 out of 16, or 75 per cent, of the animals vaccinated with either type of vaccine that finally died of generalized tuberculosis did so after 100 days. In the control group, only 3 out of 25, or 12 per cent, of the animals died more than 100 days after inoculation with H_{37}. In addition, there were 4 vaccinated guinea pigs that died of intercurrent infection 146 to 179 days after inoculation with virulent tubercle bacilli and showed little evidence of tuberculosis at autopsy.

By the same method of comparison it may be seen that 6 out of 8, or 75 per cent, of the animals immunized with the "regular" vaccine that finally died of generalized tuberculosis did so after 100 days. Exactly the same figures hold for the group vaccinated with the "bile" vaccine. Under the conditions of this experiment, there seems to be no significant difference between the immunogenic ability of the two types of vaccine.

SUMMARY

An experiment is described designed to determine whether the immunogenic ability of BCG, which is ordinarily cultured intermittently in the presence of ox-bile, is changed by continuous association with ox-bile. The results suggest that the possible loss of potency that could theoretically accrue to the vaccine because of exposure to bile has not yet made its appearance although BCG has been in continual cultivation on an artificial medium for thirty-nine years.

SUMARIO

Comparación de la Inmunogenicididad para el Cobayo del BCG Cultivado Intermitente y Continuamente en Presencia de Bilis

El experimento descrito se proponía determinar si la continua asociación con bilis de buey hace variar la capacidad inmunógena de BCG, que se suele cultivar intermitentemente en presencia de dicha bilis. El resultado denota que todavía no ha aparecido la pérdida de potencia que podria teóricamente aparecer en la vacuna debido a la exposición a la bilis, aunque BCG ha estado en cultivo continuo en medio artificial durante 39 años.

NOTE. Since the above article was submitted for publication, there has appeared the report of F. van Deinse (Am. Rev. Tuberc., 1948, *58*, 571) which confirms the experimental results reported here.

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AMERICAN TRUDEAU SOCIETY

Medical Section of the National Tuberculosis Association

STREPTOMYCIN IN THE TREATMENT OF TUBERCULOSIS

Report of Clinical Subcommittee

Committee on Medical Research and Therapy

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Pulmonary Tuberculosis

Streptomycin has a limited but important place in the therapy of pulmonary tuberculosis. In general, streptomycin should be withheld if other satisfactory treatment is available.

Streptomycin therapy is not advisable in: (a) chronic fibroid or fibro-caseous pulmonary tuberculosis; (b) acute destructive and apparently terminal types of tuberculosis, except briefly and intermittently for purely symptomatic relief; (c) the minimal stages of pulmonary tuberculosis with a good prognosis, until attendant toxicity and incidence of resistant bacteria are more precisely defined.

Streptomycin appears to be most effective in the treatment of recent, acute, fairly extensive, and progressing pulmonary tuberculous lesions. Its use is particularly recommended in the treatment of tuberculous pneumonia.

Symptomatic relief and roentgenologic improvement are common in many types of pulmonary tuberculosis soon after initiating therapy with streptomycin. In general, the degree of benefit is in direct proportion to the extent of the acute or exudative component of the total disease being treated. As relapse is common, streptomycin is best used as an adjunct to other standard forms of treatment.

The following types of disease are regarded as especially suitable for streptomycin therapy: (a) acute bronchopneumonic tuberculosis with severe symptoms; (b) acute bronchogenic spread, particularly when of such a nature that much needed collapse therapy is prevented; and (c) chronic disseminated finely nodular tuberculosis without large confluent areas of destructive disease.

As streptomycin can only be used for periods of two to four months in the therapy of this chronic disease because of the emergence of resistant organisms, it is imperative that streptomycin be used for a period during the course of the disease when the greatest benefit can be expected.

The immediate benefits of streptomycin are usually limited to a period varying from several weeks to, at most, three months. Thus it is extremely important that the period of chemotherapy be worked into an over-all plan of treatment which frequently will include collapse therapy and generally includes institutional care.

The Committee looks with disfavor on the practice of utilizing streptomycin prior to institutional care or as an alternative to collapse therapy in the hope that such treatment might be avoided.

Ulcerating Tuberculous Lesions of Mucous Membranes

Ulcerating lesions of mucous membranes usually respond well to streptomycin therapy. Included in this group are laryngeal, tracheal, bronchial, oropharyngeal, and enteral ulcerations, and tuberculous otitis media, and streptomycin is recommended for these conditions.

Streptomycin is most effective in tuberculous lesions of the larynx and tracheobronchial tree which are ulcerating; somewhat less so but still effective in lesions which are granulomatous; and still less effective in diffuse inflammatory lesions. It appears to be ineffective in purely cicatricial lesions. Other routes of administration, topical, aerosol, oral, are not sufficiently effective to be recommended as alternatives to parenteral administration.

With parenteral administration healing of ulcerative lesions is prompt. Streptomycin therapy may safely be discontinued soon after healing by epithelialization has been observed to occur (restitution of normal mucosal pattern roentgenologically in the case of tuberculous enteritis). In cases with an ultimately poor prognosis because of associated extensive pulmonary tuberculosis, streptomycin may be used for short and intermittent periods of time for its palliative clinical effects in treatment of clinical complications.

Tuberculous Sinuses and Fistulae

Streptomycin is recommended in the treatment of draining tuberculous sinuses and fistulae and appears to be highly effective in closing the infected tracts, at least temporarily, in a majority of cases, regardless of the underlying tuberculous disease.

For more permanent results in most instances, however, streptomycin is merely an excellent adjunct to suitable surgery, superior results being obtained only with this combined treatment. Such surgical measures will include wide excision or incision and drainage, depending on the location and nature of the underlying tuberculous pathology and are best instituted prior to, or at the time of, institution of streptomycin therapy.

Genito-urinary Tract

Streptomycin is recommended in the treatment of tuberculosis of the genito-urinary tract. As a rule this will be in conjunction with other indicated therapy, especially surgery. Symptomatic relief and increase in bladder capacity are common, and the urine is sterilized in many, although only temporarily in some. Results are compromised by the fact that most clinically detected cases of renal tuberculosis are far advanced with caseation, and streptomycin is least effective in these cases.

Tuberculosis of Bone, Joint and Cartilage

Streptomycin is advised in the treatment of tuberculosis of bone, joint and cartilage, particularly in conjunction with other accepted forms of therapy. Because the effect of streptomycin therapy may be long delayed in becoming apparent, it is important to exercise discriminating judgment in the timing of the use of the drug as an adjunct to other procedures.

Tuberculous Meningitis

Intensive therapy with streptomycin, administered both parenterally and intrathecally, is imperative for the treatment of tuberculous meningitis. Intramuscular therapy should be continued for from four to six months. As indicated below, the recommended dosage for intramuscular administration is two grams a day. It is recommended that

not more than 50 mg. of streptomycin be administered intrathecally every second or third day for the greater part of the period of treatment, although this amount may be given daily for the first five to seven days.

Although neurologic complications arising as a result of this regimen are frequent and may be serious, complete clinical remission in an appreciable number of cases is observed. The response is best when diagnosis is early and treatment prompt so that bacteriologic confirmation need not be secured prior to institution of therapy.

To avoid the dangers inherent in treating a nontuberculous meningitis with streptomycin, it is wise not to institute therapy with streptomycin unless at least one of the following conditions is met: (a) an associated active tuberculous focus is present with a positive tuberculin test, or (b) cerebrospinal fluid cultures are negative for ordinary pathogens and the characteristics of the spinal fluid are in keeping with those of tuberculous meningitis.

Miliary Tuberculosis

Streptomycin therapy is indicated in the treatment of both acute hematogenous (miliary) tuberculosis and chronic hematogenous disseminated tuberculosis.

Good clinical judgment is needed to differentiate these conditions from nontuberculous pulmonary infiltrations simulating miliary tuberculosis, especially in the early stages. Despite this difficulty, early treatment is highly desirable and in appropriate instances should be started even before bacteriologic confirmation is obtained. Bone marrow biopsy and culture of material for acid-fast bacilli may be a useful aid in early diagnosis. Treatment should be continued for at least four months and one to two grams a day is adequate dosage.

Development of tuberculous meningitis is a common hazard in the course of acute miliary tuberculosis. It may occur during or shortly after therapy with streptomycin. Because the onset of this complication is frequently insidious, it is recommended that routine lumbar punctures be performed at intervals during the six to eight months after the diagnosis of miliary tuberculosis. Intrathecal therapy should be added whenever the cerebrospinal fluid is found abnormal before the advent of clinical evidences of central nervous system infection.

Tuberculous Lymphadenitis

Although there is some doubt as to whether streptomycin should be employed in the treatment of tuberculous lymphadenitis, as in the case of minimal pulmonary tuberculosis (see above), it may be employed in the treatment of lymphadenitis, especially in the acute stage. It is probable that the majority of such lesions treated with streptomycin will regress or disappear promptly, although relapse may occur.

Tuberculous Peritonitis and Pericarditis

Streptomycin is recommended in the treatment of tuberculous peritonitis, for clinical remission is common and relapse relatively infrequent following adequate therapy. Streptomycin may not be expected to alter the sequelae of tuberculous pericarditis but appears to have a beneficial effect on the acute process itself.

Skin and Ocular Tuberculosis

More extensive observations will be required to evaluate the effect of streptomycin in the therapy of tuberculosis of the skin and the eye.

Streptomycin and Surgical Procedures

Streptomycin appears to be beneficial when employed prophylactically for short periods of time in association with certain surgical procedures. This is particularly true for single-stage operations, where prolonged administration of streptomycin is not required. Employed for short periods before and after operation, streptomycin has markedly decreased the hazards of pulmonary resection for tuberculosis by diminishing the incidence of early postoperative spreads.

It is not considered advisable to employ streptomycin as a routine prophylactic in multiple-stage operations for tuberculosis.

Dosage and Duration of Streptomycin Therapy

Much remains to be determined as to the optimal dosage and duration of streptomycin therapy. It is probable that no single streptomycin regimen is suitable for all types and forms of tuberculosis. With smaller daily doses of streptomycin, there is relatively little danger of toxicity; but the problem of the emergence of drug-resistant strains of organisms remains unsolved.

One gram a day may be sufficient for the majority of tuberculous lesions, with the exception of tuberculous meningitis and miliary tuberculosis, where more may be advisable (see above). There is some evidence that some regimens of two grams a day produce better therapeutic results. The higher incidence of toxic manifestations on the regimens using the larger dosages must be balanced against their possibly greater efficacy.

Frequency of administration need not be greater than every twelve hours. It is advised that streptomycin therapy be administered in courses of forty-two days (which may be repeated, if necessary) to reduce the incidence of emergence of drug-resistant organisms. This recommendation does not apply in the case of miliary tuberculosis and tuberculous meningitis, where, as noted above, it is recommended that treatment be continued for at least four months.

In individual cases it may be necessary to weigh the therapeutic advantages that might accrue from longer periods of treatment against the disadvantage of the emergence of drug-resistance. Until further studies have been concluded, it cannot be recommended that injections be made less frequently or that treatment be continued for shorter periods of time than here indicated.

Toxic Manifestations of Streptomycin Therapy

Toxic manifestations are relatively infrequent on the dose recommended above, *i.e.*, one gram a day. Nevertheless the following reactions do occur and whenever treatment with streptomycin is contemplated, the dangers of the untoward side reactions should be compared with the hazards of the tuberculous disease being treated:

(a) A disturbance of vestibular function may be observed, especially following prolonged treatment with larger doses. Partial or complete compensation is frequently noted, especially in younger persons, but the potentialities of this disorder must not be underestimated by the physician. It has not been determined if this disadvantage to streptomycin therapy can be overcome.

(b) Deafness may be produced in very rare instances and only following larger doses or when streptomycin excretion is defective. Useful hearing is nearly always regained if treatment is suspended promptly when deafness is noted. Audiometric observations are probably advisable at this time until the conditions under which deafness occurs are better defined.

(c) Serious renal damage produced by streptomycin appears to be observed rarely except when there is pre-existing renal disease.

(d) Cutaneous rashes, apparently due to acquired hypersensitivity to streptomycin, are occasionally observed and sometimes indicate that treatment should be suspended temporarily. It is usually possible to resume treatment later. Serious exfoliative dermatitis is observed rarely.

Emergence to Predominance of Drug-resistant Tubercle Bacilli

The disappearance of drug-sensitive strains of tubercle bacilli and their replacement with strains which are drug-resistant present a serious handicap to prolonged effective therapy with streptomycin. It is not yet known what conditions encourage the appearance of drug-resistant organisms, how to determine drug-resistance with absolute precision, or how permanent this change in bacterial flora may prove to be. There appears to be a fairly uniform rate of appearance of such drug-resistance, related to the duration of treatment but independent of the daily dosage. The advantage of the forty-two day duration of therapy suggested is the probable avoidance of the emergence of drug-resistant strains in a larger percentage of cases. Retreatment is possible and feasible if the organisms remain drug-sensitive and the disease continues an unfavorable course or relapse occurs. Organisms should be tested for sensitivity before retreatment is undertaken, whenever practicable.

Dihydrostreptomycin

The Committee has reviewed limited experimental and clinical evidence concerning dihydrostreptomycin, a hydrogenated derivative of streptomycin. In most respects, the pharmacological properties of this compound, including absorption and excretion, are similar to those of the parent substance and in comparable doses dihydrostreptomycin and streptomycin apparently produce the same suppressive action on the tubercle bacillus, *in vitro* and *in vivo*.

The chief advantage of dihydrostreptomycin is that it is less toxic than streptomycin, especially in its action on the vestibular apparatus, when given in comparable doses for similar periods.

Despite the lowered incidence of toxicity, it should be pointed out that dihydrostreptomycin is not devoid of toxic potentialities. In addition to neurotoxicity, abdominal discomfort, nausea, vomiting, and skin rashes have occurred in a few instances. Another possible advantage of this compound is that it appears to be well tolerated by some patients who have developed hypersensitivity reactions to streptomycin. These advantages seem promising and warrant extensive clinical trials.

Some preparations of dihydrostreptomycin have produced local irritation at the site of injection; such preparations should not be used intrathecally. Unfortunately, strains of tubercle bacilli resistant to dihydrostreptomycin emerge in the same manner as to streptomycin, and organisms resistant to one compound are equally resistant to the other.

AMERICAN TRUDEAU SOCIETY

Postgraduate Courses in Pulmonary Diseases

The following postgraduate courses in pulmonary diseases are scheduled for the first half of 1949 by the American Trudeau Society, Medical Section of the National Tuberculosis Association. This program is conducted under the auspices of eight regional committees, covering all states, and the provinces of Canada, in cooperation with the medical schools of leading universities.

Applications may be obtained from the American Trudeau Society, 1790 Broadway, New York 19, New York. These courses are generally oversubscribed and physicians interested in applying are urged to request application forms as early as possible.

By special arrangement with the Department of Medicine and Surgery of the Veterans Administration, physicians employed by that agency are invited to participate. The request for detail to these courses originates in the branch and regional offices of the Veterans Administration and duplicate applications are required.

Physicians applying under Public Law 346, (G I Bill) should so state when filing applications with the Society.

January 24–29, 1949 Los Angeles, California	Region VII comprising the states of: Washington, Oregon, California, Idaho, and Nevada; and the provinces of Alberta and British Columbia, Canada. This one week course will be held in cooperation with the College of Medical Evangelists, the University of Southern California School of Medicine, and the University of California at Los Angeles Medical School. Tuition—$50.00
March 7–12, 1949 Indianapolis, Indiana	Region V comprising the states of: Ohio, Indiana, Michigan, Illinois, Wisconsin, Missouri, Iowa and Minnesota; and the provinces of Manitoba and Saskatchewan, Canada. This one week course will be held in cooperation with Indiana University School of Medicine. Tuition—$50.00
March 13–26, 1949 New Orleans, Louisiana	Region IV comprising the states of: Alabama, Arkansas, Louisiana, Missippi, Oklahoma and Texas. This two week course will be held in cooperation with Tulane University of Louisiana School of Medicine and Louisiana State University. Tuition—$100.00
April 4–9, 1949 Atlanta, Georgia	Region III comprising the states of: Maryland, Virginia, West Virginia, Kentucky, Tennessee, North Carolina, South Carolina, Georgia, Florida and the District of Columbia. This one week course will be held in cooperation with Emory University School of Medicine. Tuition—$50.00
July–August, 1949 Denver, Colorado	Region VI comprising the states of: Colorado, North Dakota, South Dakota, Nebraska, Kansas, New Mexico, Arizona, Utah, Wyoming and Montana. This two week course will be held in cooperation with the University of Colorado School of Medicine. Tuition—$100.00

Spring or Summer, 1949	Region VIII is planning a course at Saranac Lake, N. Y., chiefly for physicians from Northern New York and the Canadian Province of Quebec. This will be held in cooperation with medical schools of the area and The Trudeau Sanatorium. The dates have not yet been announced but information will be sent to those requesting it when available.

Special attention is directed to the following course for general practitioners which is being given in cooperation with The St. Louis University School of Medicine, January 17, 18, 19, 1949. Applications for this course should be obtained from the Executive Secretary of the Missouri Trudeau Society, 411 North 10th Street, Room 505, St. Louis 1, Missouri.

This is the second course for general practitioners included in the American Trudeau Society's program of postgraduate opportunities. There follows a summary of some of the subjects to be presented by leaders in the field of pulmonary disease:

Symptomatology in Chronic Pulmonary Disease—Practical evaluation of cough, pain, hemoptysis and dyspnea. Julius L. Wilson, M.D., Professor of Clinical Medicine, Tulane University School of Medicine; Head of Section on Chest Diseases, Ochsner Clinic, New Orleans, La.

Physical Examination of the Chest—its limitations. Paul Murphy, M.D., Assistant Professor of Clinical Medicine, St. Louis University School of Medicine, St. Louis, Mo.

Chest Films—how to obtain satisfactory ones. Don C. Weir, M.D., Senior Instructor in Radiology, St. Louis University School of Medicine, St. Louis, Mo.

Skin Tests—their importance in the diagnosis of lung diseases. Herbert L. Mantz, M.D., Tuberculosis Controller, Kansas City, Mo., Consultant in Tuberculosis, Veterans Administration.

The Five Cardinal Points in the Diagnosis of Clinical Pulmonary Tuberculosis. George D. Kettelkamp, M.D., Medical Director, Robert Koch Hospital; Assistant Professor of Clinical Medicine, Washington University School of Medicine, St. Louis, Mo.

Basic Concepts and Objectives in the Treatment of Pulmonary Tuberculosis. Carl Muschenheim, M.D., Associate Professor of Clinical Medicine, Cornell University Medical College, New York, N. Y. (Dr. Muschenheim also will speak on "Streptomycin in the Treatment of Tuberculosis.")

Among other subjects to be presented are: extra pulmonary tuberculosis, special problems in the management of tuberculosis such as pregnancy, diabetes, syphilis, surgical operations; tuberculous pleurisy with effusion, its management from the standpoint of avoiding subsequent phthisis; public health aspects of pulmonary tuberculosis, mass X-ray surgery, tuberculin testing, general hygiene; the pneumonias, practical points in the diagnosis and management; bronchiectasis; empyema thoracis; bronchogenic carcinoma; mediastinal tumors; mycotic diseases of the lung; the pneumoconioses; use of oxygen therapy, and rare diseases of the lung.

THE

AMERICAN REVIEW OF TUBERCULOSIS ABSTRACTS

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Streptomycin for Tuberculous Meningitis.—Clinical trials of streptomycin in the treatment of tuberculous meningitis in Great Britain were conducted under the supervision of a committee of the Medical Research Council. The results in 105 cases are reported. Of these, 71 have died, 33 are making good progress and one is deteriorating after seven or more months of treatment and observation. The prognosis in children under 3 years of age was much worse than in older children. The prognosis was better in the early stages of the disease than in the later. The results in patients who received only intramuscular streptomycin were much poorer than in those receiving the drug both intramuscularly and intrathecally. Nevertheless, it was felt that daily intrathecal treatment sustained over a long period ceases to be beneficial and probably is harmful. It was felt also that periods of complete rest from streptomycin treatment are beneficial. The streptomycin levels in the cerebrospinal fluid were found to be lower in patients who were recovering than in those who were deteriorating. Among the fatal cases 4 types were noted: (*1*) no response to treatment, (*2*) slow progressive deterioration with no period of improvement, (*3*) progressive deterioration after a short initial period of improvement or no change, and (*4*) relapse after a long period of improvement. In all but the first group, life was prolonged far beyond the normally expected period. As a result of the longer period of survival, spinal block and hydrocephalus were found in greater frequency than heretofore. In patients who ultimately fared badly, tubercle bacilli were isolated from the cerebrospinal fluid much more frequently during the first three weeks in patients who did poorly than in those who made good progress. Strains isolated from 22 patients treated up to 136 days were found resistant to streptomycin in only 3 cases.—*Streptomycin Treatment of Tuberculous Meningitis, Medical Research Council, Lancet, April 17, 1948, 1: 582.*—(*A. G. Cohen*)

Streptomycin for Tuberculous Meningitis.—A total of 18 cases of tuberculous meningitis were treated with streptomycin. Of these, 7 died and 11 survived; 4 of the latter are still under treatment. In most cases, the drug was administered both intramuscularly and intrathecally for at least six weeks and thereafter intramuscularly alone for the next four to six months. The average intramuscular dose was 2 Gm. daily for adults and 0.02 Gm. per lb. of body weight for infants, at first by divided doses and then by a single daily dose. After about four months, if the patient was doing well, a single injection of 2 Gm. was given every two days. The daily intrathecal dose was 0.1 Gm. for adults and 0.05 to 0.07 Gm. for infants, given as a single injection. When very strict antiseptic precautions were used, very few side effects were seen, except involvement of the eighth cranial nerve and gastric upsets. In the fatal cases, necropsy revealed extensive exudative changes in the meninges. These resulted in hydrocephalus, symptoms of cerebral anoxia or infection, and manifestations of hypothalamic disturbance. The diagnosis of ad-

vanced cases was relatively simple; it was easy to find acid-fast bacilli on smears and to confirm their pathogenicity by culture. In early cases, the diagnosis sometimes could not be proved immediately and treatment was begun in some cases before the organisms were identified. In all but 2 cases, frontal burr holes were made to provide access to the anterior horns of the lateral ventricles. If 7 to 10 ml. of fluid were obtained from each side, the diagnosis of brain abscess could be excluded and treatment begun earlier. Sometimes the organism was found more readily in the ventricular specimen than in the lumbar. Direct access to the ventricles for administration of streptomycin was thus provided. The object of treatment was to obtain as soon as possible a high level of streptomycin in the cerebrospinal fluid and to maintain this level. In the 7 survivors who have stopped treatment, neurological recovery is complete except for the eighth nerve.—*Treatment of Tuberculous Meningitis with Streptomycin, H. V. Smith, R. L. Vollum and H. Cairns, Lancet, April 24, 1948, 1: 627.*—(*A. G. Cohen*)

Streptomycin for Tuberculous Meningitis.—About 75 cases of spontaneous recovery from tuberculous meningitis have been reported in the literature. No case has ever recovered, however, in the past thirty-two years at the Boston City Hospital; the deaths have totalled 178. A 7-year-old boy has been treated with streptomycin and is clinically cured after one year. He received a total of 70,458,332 units intramuscularly and 2,675,000 intrathecally over a period of 141 days. He has several residual paralyses. Two other patients died after brief treatment; a third infant recovered clinically, was sent home with serious cerebral damage, and died thirteen months after the start of therapy; a fourth child, apparently completely well, died suddenly nine months after she was first seen, and a cerebral thrombosis and infarction (produced by the growth of a tuberculoma) were found, as well as miliary tubercles containing acid-fast bacilli. Early and intensive therapy is recommended.—*Tuberculous Meningitis Treated With Streptomycin, O. S. Nau, Jr. & F. J. Wenzler, J. Pediat., April, 1948, 32: 410.* (*W. H. Oatway, Jr.*)

p-Aminosalicylic Acid for Tuberculosis.—Five patients with pulmonary tuberculosis were treated with p-Aminosalicylic Acid for sixty days and one patient for four weeks. A total of 12 Gm. daily was administered in divided doses every three hours. Within a few days there was great improvement in the patient's general condition. There was a decline in fever and a decrease in the number of organisms. A blood level of 2 to 5 mg. per cent was reached with the stated dose. There were no toxic reactions. The author believes that the dosage and length of treatment were insufficient but that the drug is of value in exudative and toxic forms of tuberculosis.—*Pulmonary Tuberculosis Treated with p-Aminosalicylic Acid. A. Erdei, Lancet, May 22, 1948, 2: 791.*—(*A. G. Cohen*)

Whooping-cough and Tuberculosis.—In the ten years before August 1947, 1,747 patients were admitted to the Cleveland City Hospital with pertussis. All such patients routinely are tuberculin skin-tested, since the pulmonary symptoms of tuberculosis may be the same as those of pertussis and the findings by X-ray may be similar for several infections. In 38 patients the chest X-ray showed suspicious evidence of tuberculosis; in 23 patients the tuberculin test was positive; 19 actually had either primary, active, or arrested tuberculosis. Except in one case there was no evidence that tuberculosis was activated by pertussis. All patients ill with whooping-cough should have a tuberculin skin test to aid differential diagnosis.—*Does Pertussis Activate Tuberculosis?, J. A. Toomey, J. C. Berno, & H. Agustsson, J. Pediat., March, 1948, 32: 260.*—(*W. H. Oatway, Jr.*)

Coexisting Coccidioidomycosis and Tuberculosis.—Only 3 such cases are recorded in the literature. There have been recent verbal reports of several more. The authors report a case of cavitation due to coccidioido-

mycosis. After fifteen months, the patient developed cavitary tuberculosis in the opposite lung.—*Coexisting Pulmonary Coccidioidomycosis and Tuberculosis, R. S. Study & P. Morganstern, New England J. Med., June 10, 1948, 238: 887.*—(*A. G. Cohen*)

Serous Tuberculosis in African Negroes.—Among native African troops between the ages of 18 and 30, 169 cases of serous pleuritis were seen. These are divided as follows: primary glandular tuberculosis with effusion, 16, primary unilateral effusion, 125, and tuberculous polyserositis, 28 cases. The 16 cases of primary glandular tuberculosis with effusion are part of a group of 36 cases of primary glandular tuberculosis. The patients gave a history of fever, cough and slight expectoration for several weeks. Some showed no local signs or symptoms and were first regarded as cases of unexplained fever. Others showed signs of local consolidation and were first regarded as pneumonia. X-rays showed, besides the effusion, enlarged nodes and occasionally hilar flare or definite lobar or segmental collapse. Sputum and gastric studies were negative for tubercle bacilli. The effusion appeared at the onset in 7; in the other 9 it appeared after three to four months. The course in all patients was benign; there were no deaths. The temperature gradually fell and the fluid absorbed but the glandular enlargement persisted. The second group consists of 125 cases of primary pleural effusion with no deaths. The onset in some was subacute with chest pain, fever and dry cough. In others, the onset was insidious with general malaise and minimal local symptoms. In others, there was an acute onset with high fever and severe pleuritic pain; these cases were diagnosed as pneumonia at first. In 12 per cent, there was enlargement of the supra-clavicular, less often the cervical, lymph nodes. In the few cases checked, cultures of the fluid were negative. The cells were predominantly lymphocytes. The sputa were negative. The white blood counts were normal. The subsequent course was characterized by a fall in temperature and gain in weight. There was a gradual fall in sedimentation rate; higher final figures were found in cases with the most residual pleural thickening. It was impossible to enforce bed-rest after the initial acute stage. In about one half of the cases the fluid was allowed to absorb without interference; in the other half, it was aspirated as indicated. The final results showed considerable pleural thickening in 70 per cent; there was little difference between the 2 groups. Criteria for arrest were: (*1*) normal temperature for three weeks, (*2*) absorption of fluid, (*3*) significant weight gain, and (*4*) progressive fall in sedimentation rate. This required about four to five months. The third group consists of 28 cases of polyserositis. These are divided as follows: (*1*) presenting as primary glandular tuberculosis, with subsequent generalized miliary spread, 3 cases, all fatal; (*2*) primary serous tuberculosis, with subsequent consecutive effusions of the polyserositis type, 11 cases with one death; and (*3*) primary serous tuberculosis with subsequent polyserositis, 14 cases with one death. In these 28 cases, 20 presented as a unilateral pleural effusion; a contralateral effusion appeared within eighteen months in 15. Pericardial effusion was found in 8 and abdominal glandular and serous tuberculosis in 10 cases. Anatomically, all the cases were characterized by marked tracheobronchial lymph node tuberculosis. The disease then progressed from one chain of glands to the other and may have been the cause of the pleural effusion. Unlike the case in Europeans, primary pleural effusion in African Negroes is linked with slow continuous or intermittent evolution of primary tuberculosis. It appears in the post-primary stage within about six months and displays 3 tendencies unusual in European adults: (*1*) it is an incident in a predominantly glandular syndrome; (*2*) it may develop into polyserositis; and (*3*) at any point, miliary dissemination may supervene.—*Serous Tuberculosis in East African Natives, S. R. Wood, Brit. J. Tuberc., April, 1948, 42: 38.*—(*A. G. Cohen*)

Congenital Tuberculosis.—A previous definition of congenital tuberculosis has been "tuberculosis in which the infection occurs before birth by way of the blood stream or by aspiration of the amniotic fluid or contents of the birth canal." The authors agree with the first portion, but believe that those cases which result from aspiration in the birth canal should be called "tuberculosis neonatorum." Several reviews of the literature in recent years have "authenticated" 100 cases, more or less, and some believe that intra-uterine infection is quite rare. The authors report the case of a Negro infant, removed from its home within an hour after a normal birth, which died on the forty-fourth day of acute miliary tuberculosis, proved at necropsy. The mother died sixty seven days post partum of a "miliary tuberculosis," diagnosed by X-ray of the chest. She also had a positive Wassermann, a negative tuberculin skin test, and a foul vaginal discharge.—*Congenital Tuberculosis. A Review of the Disease With Report of a Case, E. A. Harris, G. C. McCullough, J. J. Stone, & W. M. Brock, J. Pediat., March, 1948, 32: 311.*—(*W. H. Oatway, Jr.*)

Extrapleural Pneumolysis with Lucite Plombage.—There has been a continuous search for the ideal substance to fill the extrapleural space in plombage. Certain experimental work suggested that methyl methacrylate, known commercially as lucite, was well suited for this purpose. Balls of this substance were used in operations on 30 cases. The lesions were of the same type as those for which thoracoplasty would ordinarily be performed except that those with large thin-walled cavities were not taken. Lesions down to the third anterior rib were considered operable. Advanced age and bilaterality were not considered contraindications. All were pneumothorax failures. An average of 20 to 35 lucite balls were used in each operation. The postoperative course generally was uneventful. All patients developed fluid in the extrapleural space. The febrile reaction was mild. Subcutaneous emphysema was infrequent. The pleura was torn in 3 cases but with no serious outcome. Most patients had some pain. The advantages of the operation are lack of deformity, absence of paradoxical motion and less loss of pulmonary function. An economic advantage is the fact that it is performed in one stage. It is a more difficult operation than thoracoplasty. The ultimate fate of the lucite is unknown.—*Extrapleural Pneumonolysis with Lucite Plomage, J. B. Grow & R. E. Dwork, Brit. J. Tuberc., April, 1948, 42: 24.*—(*A. G. Cohen*)

Extrapleural Pneumonolysis. — Lateral thoracid branches of the internal mammary artery occur in approximately 50 per cent of individuals. These vessels course over the inner aspect of the upper and anterolateral area of the chest wall. They may be damaged when giving refills after an extrapleural pneumonolysis, resulting in serious hemorrhage into the extrapleural space. Moderate or massive hemorrhage occurs in 5 to 20 per cent of cases and may result in collapse of the patient or, more remotely, delayed infection or gradual obliteration of the space. When discovered at operation, the vessels should be ligated or coagulated with diathermy and, in addition, refills should at first be given from the back.—*Hemorrhage after Extrapleural Pneumonolysis, W. P. Cleland, Thorax, June, 1948, 3: 127.*—(*A. G. Cohen*)

Bilateral Collapse Therapy.—In a group of thoracoplasty cases, 11.2 per cent received contralateral pneumothorax. These were divided into 3 groups according to the postoperative results. Group A, comprising those with negative sputum, totaled 14 cases or 48.25 per cent. In all cases the lesions were arrested or quiescent. In group B, with positive sputum, there were 10 cases or 34.5 per cent. The lesions were stationary or improving in 5 and extending in 5. Group C contained 5 cases or 17.25 per cent; these were all dead. The initial prognosis in these cases, requiring thoracoplasty in the presence of an active contralateral lesion, is bad. The

pneumothorax must be managed with the greatest care. A marginal pneumothorax should be maintained and an attempt should be made to keep the lung at a constant distance from the chest wall. Three of 7 relapses and 2 of the 5 deaths resulted directly from premature abandonment of the pneumothorax. The optimum time for such abandonment is debatable. The author believes it should be maintained as long as possible.—*Bilateral Active Pulmonary Tuberculosis Treated with Artificial Pneumothorax and Thoracoplasty, G. P. Maher-Loughann, Tubercle, May, 1948, 29: 101.—(A. G. Cohen)*

Recurrent and Chronic Spontaneous Pneumothorax.—Seventy-one cases of recurrent and chronic spontaeous pneumothorax are reported. A case is considered chronic if the collapse lasts longer than three months. There were 46 such cases with an average duration of fifteen months. Of these, 29 were entirely chronic while in 17 chronicity supervened upon recurrent attacks. There were 25 purely recurrent cases; when combined with the 17 complicated by chronicity, they make a total of 42 recurrent cases. The average number of attacks was 4; the greatest number was 15. Chronic pneumothorax leads to invalidism and disability. In recurrent pneumothorax, the patient is doomed to a life of inactivity. It is believed that chronic and recurrent pneumothorax are manifestations of one aspect of the natural history of many lung diseases. The number of males greatly exceeded females. All decades of life were represented. There was a higher incidence in older decades than in pneumothorax simplex. Clinically, there was a greater coincidence of chronic bronchitis and emphysema. The condition often was noted accidentally. In chronic pneumothorax the loss of weight was striking. Plain radiographs may show nothing more than the pneumothorax. The first consideration is to decide whether it is actually a pneumothorax or rather a giant cyst or bulla. Diagnostic puncture is dangerous as it may lead to a severe pneumothorax; it should be done only when one is prepared to proceed with thoracotomy. The plain film should be inspected for evidence of cysts or bullae. These are easier to detect by tomography. Bronchograms occasionally are desirable; they may disclose unsuspected or asymptomatic bronchiectasis and may also help in differentiating between pneumothorax and cyst. Bronchoscopy occasionally is helpful. Pleural pressure readings which quickly revert to the original after aspiration are diagnostic of a bronchopleural fistula. Determination of the carbon dioxide, oxygen and nitrogen content of the pleural air also may help to determine the presence of a fistula. The most valuable diagnostic procedure is thoracoscopy. The following types of underlying lesion were found: generalized emphysema in 12, bullous emphysema in 13, asthma and bronchitis with emphysema in 8, large solitary bullae or cystic disease in 11, diffuse polycystic disease in 3, small bullae, mostly apical, in 15, apical scars in 6, leak or tear seen in 4, areas of "cuckoo-spit" in 4, various, such as tooth extraction, staphylococcus abscess, drainage of empyema and tuberculous pleurisy each in one and no cause in 6. "Cuckoo-spit" is a name given to a condition in which there is an ooze of air bubbles from an apparently normal lung surface. Thus, the cause of spontaneous pneumothorax may lie in a defect in either the pleura, subpleura, alveoli or bronchi. Large cysts or bullae are treated surgically by excision or lobectomy. In other cases, pleurodesis is effected. The author prefers to use silver nitrate. Four swabs of a 20 per cent solution are first applied directly to the lung surface at the time of thoracoscopy. Later, 5 to 10 minims of a 10 per cent solution are injected. This results in fever and effusion. There must then be judicious aspiration of air and fluid.—*Recurrent and Chronic Spontaneous Pneumothorax, R. C. Brock, Thorax, June, 1948, 3: 88.—(A. G. Cohen)*

Welder's Siderosis.—In 1936, shadows were discovered in the chest X-rays of electric-arc welders. These men were apparently in

good health. It was believed that the shadows probably were due to deposition of iron oxide. Fibrosis does not result from this. Since then, many electric-arc, carbon-arc and oxyacetylene welders have been examined and similar shadows were found in each group. In 1945, 15 of the original subjects were re-examined; all but 2 had continued at the same occupation. Of 7 whose X-rays had shown no abnormalities, one was suspicious and one showed definite changes. In 2 with suspicious shadows, the changes had become definite. Of 6 with positive findings, 4 were still so, one appeared to be clearing and one was negative. Siderosis is one of the benign pneumoconioses in which dusts deposited in the lungs give characteristic X-ray changes but no disability. These changes are not necessarily permanent.—*Clearing of X-ray Shadows in Welder's Siderosis, A. T. Doig & A. I. McLaughlin Lancet, May 22, 1948, 2: 789.*—(*A. G. Cohen*)

Bronchiectasis.—In a military hospital, 211 men and 3 women aged 18 to 49 showed symptoms and signs resembling chronic bronchitis. Bronchography was performed for all; 46 (21 per cent) showed bronchiectasis. There was a past history of pneumonia in 26; in 6 there had been 2 or more attacks and one patient had had 4 attacks. Pleurisy alone had been present in 3 cases. There was no evidence of tuberculosis although suptum examinations had been made for about half of the cases. Four patients had had measles and one had pertussis. The most frequent previous symptom was a winter cough. Currently, the patients suffered from productive cough and dyspnea. The cough was aggravated by changes of temperature and by smoking. In most cases, it was severe enough to cause disability. The sputum was either white mucoid or thick yellow and gave no odor; about 2 to 3 ounces were produced in twenty four hours. Night sweats were present in 7 cases, loss of weight in 2 and mild hemoptysis in 12. Examination showed diminished respiratory excursions in all cases. Clubbing of the fingers was found in 8 cases. Impairment of percussion note, poor air entry and coarse rales were common. The signs and symptoms were the same in both the bronchitis and bronchiectasis group, but were more frequent in the latter. The bronchiectases as demonstrated by bronchography were cylindrical in 31, varicose in 3 and saccular in 12. Plain chest films showed increased markings which were sometimes crowded together. More rarely there were localized zones of increased translucency in the middle or outer part of the lower lobe. This sign, if found, is very reliable. Other changes described in the literature are not definitive and bronchography is essential for diagnosis. The bronchi in unresolved pneumonia may first undergo reversible mechanical dilation which may later be rendered irreversible by infection. The reversible state lasts for several months.—*Bronchiectasis Simulating Chronic Bronchitis, J. D. H. Wearing, Lancet, May 29, 1948, 1: 822.*—(*A. G. Cohen*)

Liver Biopsy in Sarcoidosis.—Aspiration liver biopsies in 3 cases which presented clinical features suggesting a diagnosis of sarcoidosis showed typical noncaseating tubercles. In one case, serial sections were necessary. A typical lesion also was found in a liver biopsy from a patient presenting no other evidence of the disease.—*Liver Biopsy in Sarcoidosis, J. G. Scadding & S. Sherlock, Thorax, June, 1948, 3: 79.*—(*A. G. Cohen*)

Prophylaxis of U.R.I.—One hundred and forty-eight children, living in a metropolitan area and susceptible to various upper respiratory infections, were given 50,000 units of buffered penicillin in tablet form before two meals a day for one year. The effect was determined by comparing the number of infections and the days of fever with those of the previous year and against those of 110 comparable controls who received no penicillin. Cultures and blood levels were not determined. The controls showed no change in the incidence of infection or days of fever. The treated group showed a decrease in the

number of infections to 45.5 per cent of the previous year, and a decrease of febrile days to 25.3 per cent. This confirms the results of a previous test in which 25,000 units per day and influenza virus vaccine were used. Only three children showed any toxic reaction; there was no evidence of a resistant bacterial flora.—*Prophylaxis of Upper Respiratory Infections in Children Treated with Oral Penicillin, J. H. Lapin, J. Pediat., February, 1948, 32: 119.—(W. H. Oatway, Jr.)*

Spontaneous Pneumothorax.—One hundred consecutive cases of spontaneous pneumothorax were studied. Of these, 64 were known to have underlying pulmonary disease. In 20 per cent, there was a history of unusual exertion prior to the episode. In 80 per cent, it occurred during ordinary activity. Tuberculosis was noted in 38 cases, emphysema in 5, bronchiectasis in 5. Empyema, asthma, abscess, carcinoma, pneumonia and infarct each occurred in a few. The great majority of the patients were males; the average age was 27 years. In 3 patients, there were multiple recurrences; one other subsequently developed spontaneous mediastinal emphysema and another developed tuberculosis after 4 recurrences. Aspiration of air was necessary in only 3 cases; in the others bed-rest sufficed. In patients over 45 years of age, the appearance of spontaneous pneumothorax should arouse suspicion of underlying carcinoma.—*Spontaneous Pneumothorax, R. M. Myerson, New England J. Med., April 1, 1948, 238: 461.—(A. G. Cohen)*

Pneumonectomy.—One of the chief complications of pneumonectomy is infection of the site previously occupied by the lung which has been removed (the gap). Various methods of obliterating the gap, including thoracoplasty, have not been successful. In the case described, pneumoperitoneum was combined with Monaldi suction applied to the gap. The Monaldi phase must await closure of the bronchial fistula. With this method, the gap was obliterated in a few months.—*Closing the Pneumonectomy Gap, G. S. Eedy, Lancet, June 12, 1948, 1: 905.—(A. G. Cohen)*

Pulmonary Hydatid Disease.—The author reviews the treatment of 27 of his own cases of pulmonary hydatic cyst. The conservative type of operation is favored. If the lung is adherent to the chest wall and the cyst is not infected, the contents of the cyst are removed and the adventitia is drained. If the cyst is infected, the cyst space is drained following removal of the contents. If the lung is not adherent, then, in addition to the above, the adventitia is sutured to the chest wall and the pleural cavity is drained. Lobectomy may be necessary for (*1*) serious hemorrhage from the cyst space during a conservative operation; (*2*) residual bronchiectasis, if symptoms warrant it; (*3*) empty sac, if hemorrhage or infection warrant it; (*4*) an uncertain diagnosis; and (*5*) giant cyst. In the author's series, 23 patients were operated upon, with one death; 4 patients coughed up their cysts.—*The Treatment of Pulmonary Hydatid Disease, M. P. Susman, Thorax, June, 1948, 3: 71.—(A. G. Cohen)*

Aspiration of Timothy Grass.—Eight cases of aspiration of timothy grass heads were seen in patients ranging from 14 months to 14 years of age. A definite history of aspiration was obtained in 5 patients; all had shown immediate symptoms which were sustained and progressive. The early symptoms, fever, cough and weight loss, led to a variety of diagnoses. The possibility of foreign body was suspected in only 2 cases. Among late symptoms, there was intermittent fever despite chemotherapy. Foul sputum was found in 6 cases, massive hemoptysis in 2, and weight loss in 3. Clubbing of the fingers was not prominent. The chest X-rays, when finally made, were diagnosed as pneumonitis, atelectasis, pneumonia or abscess. Bronchiectasis was demonstrated by bronchography in 5 cases; in addition, multiple small abscesses were seen in one of these. Bronchoscopy was performed in 7 cases. The timothy head was recovered in 3 cases; it was not found in the other 4. In one case, complete recovery followed removal. The foreign body was retained from 2 to 36 months and

symptoms appeared 4 to 36 months prior to lobectomy in the others. The right lower lobe was resected in 6 cases and the left lower lobe in one. The postoperative course was uneventful in all but one case in which there was unexplained fever for several weeks. The patients have been well for two months to four years. There were no specific pathological changes due to timothy *per se*. Three basilar abscesses containing timothy were found. Of 4 cases of classical bronchiectasis, timothy heads were found in the ectatic bronchi in 2; the stem always pointed down and the barbules upwards. The most striking histological change was extensive connective tissue proliferation about the bronchi and vessels with extension along the pulmonary septa; this was most marked in the younger children. In the older children, localized abscess formation was more prominent.—*Bronchiectasis Following Aspiration of Timothy Grass, M. G. Carter & K. J. Welch, New England J. Med., June 10, 1948, 238: 832.* —(*A. G. Cohen*)

Mediastinal Bronchiogenic Cysts.—The vast majority of mediastinal cysts are congenital in origin. Those arising from the respiratory system are termed bronchiogenic; those from the digestive tract are called gastric or esophogeal cysts. There is no sharp line of distinction. The current discussion excludes intrapulmonary cysts. The increasing use of chest X-rays has revealed more than the rare cases formerly seen, and it now can also be said that they are sometimes symptomatic. Symptoms usually begin in adult life, when the cyst has grown enough to produce pressure on adjacent structures. Eight cases are reported in this series, the literature is reviewed, and collected cases are analyzed. Bronchiogenic cysts may be classed as paratracheal, carinal, hilar, paraesophageal, or miscellaneous. In the absence of infection, symptoms depend on the size and location of the mass. Symptoms, differential diagnosis, occurrence, and treatment are discussed. Because of the congenital origin, one must bear in mind the possible presence of other anomalies. In the several reports during the past few years, a summary of surgical results has been difficult, due to incomplete diagnosis, incomplete excision, the use of aspiration, et cetera. A posterolateral transpleural approach is recommended and other surgical suggestions are given. Of the 8 patients reported, 7 had an uneventful recovery, while the eighth recovered after the effects of pericardial manipulation and a flare-up of hyperthyroidism had subsided. Surgical excision is usually recommended for the condition even though asymptomatic when first discovered. —*Bronchiogenic Cysts of the Mediastinum, H. C. Maier, Ann. Surg., March, 1948, 127: 476.*—(*W. H. Oatway, Jr.*)

Treatment of Empyema.—Twenty patients with empyema were treated by aspiration of the pus and instillation of either penicillin alone or with other agents. Six cases were caused by pneumococci, 9 by streptococci, one by staphylococci, and the others by miscellaneous organisms. In 8 cases, the pus had a putrid odor. Cures were obtained in 16, while 4 required surgical drainage. The average time of treatment was fourteen days, the number of aspirations 7.7 and the daily dose of penicillin 63,250 units. Systemic penicillin also was given. No chronic empyemas developed. Complete evacuation of the pleural fluid was done daily, followed by instillation of 100,000 units of penicillin plus 1 Gm. of streptomycin or 2 to 4 Gm. of a sulfonamide drug, depending upon the sensitivity of the bacteria. This was continued until the cultures were sterile, during which time the walls of the cavity were kept apart. The space was then obliterated as quickly as possible by aspiration of liquid and gas every other day, supplemented by penicillin instillation. Causes of failure were: (*1*) inability to evacuate the cavity because of thick pus, (*2*) nonsusceptible organism, (*3*) inability to maintain antibiotic fluid in the cavity (large bronchopleural fistula), (*4*) multiple loculi and (*5*) recurrence.—*The Treatment of Empyema With Topical and Systemic Penicillin and Other Antibacterial Agents, W. E. Burnett,*

G. P. Roscmond, J. H. Hall & H. T. Caswell, Surg., Gynecol. & Obstet., July, 1948, 87: 44.—(A. G. Cohen)

Calcification in the Lungs.—A review of the discovery, distribution, and occurrence of calcification in the lungs has been made. A series of 13 Chicago children is reported whose chest X-rays showed calcification and whose tuberculin skin tests were negative. They all appeared normal but 8 had chronic cough and frequent respiratory infections. Bronchoscopy showed a roughening of the mucosa in the main bronchi but bacteriologically negative secretions. Twelve were skin-tested with histoplasmin, and 6 were immediately positive.—*Nontuberculous Pulmonary Calcification, A. D. Biggs & R. G. Rigby, J. Pediat., April, 1948, 32: 898.—(W. H. Oatway, Jr.)*

Anomalous Left Middle Lobe.—Accessory lobes of the lungs are not uncommon, the azygos lobe being the most frequent. The retrocardiac lobe at the right base is demonstrated radiologically in 0.5 per cent to 8 per cent of chest X-rays. The lingula of the left lung is the homologue of the right middle lobe and a fissure separating the upper from an anomalous middle lobe on the left has been described. Sante found this occurred in one of 5,000 autopsies. Chest X-rays of 3 patients showed a fine line running from the left hilum toward the periphery at about the level of the posterior end of the eighth rib. The editor, incidentally, mentions 2 additional cases of his own that show this fine fissure on the left.—*The Anomalous Middle Lobe of the Left Lung, G. L. Hardman, Brit. J. Radiol., February 1948, 21: 70.—(B. Hyde)*

Resistance Breathing.—When dogs anesthetized with nembutal were made to breathe against an airway resistance of 20 cm. of water during either inspiration or expiration, significant degrees of pulmonary edema, congestion, and hemorrhage were observed upon sacrifice at six to eight hours or at the time of spontaneous death. Bilateral cervical vagotomy did not increase the tendency to lung edema under these circumstances. Pulmonary lesions after resistance breathing appear to depend upon multiple factors and cannot at present be accounted for in terms of simple direct effects of pressure upon the pulmonary tissues and vascular bed. An analysis of actual mechanisms awaits acquisition of additional factual data.—*Observations on the Effects on the Lungs of Respiratory Air Flow Resistance in Dogs with Special Reference to Vagotomy, S. Zinberg, G. Nudell, W. G. Kubicek, & M. B. Visscher, Am. Heart J., May, 1948, 35: 774.—(G. C. Leiner)*

Atelectasis in Tuberculosis.—The authors deal with the question of involvement of the peripheral bronchi in cases of so-called opaque lobe occurring during pneumothorax. In 26 necropsies on cases with tension cavities the major bronchi to the affected area were found to be involved specifically in 83 per cent of cases. The chief changes, however, were in the surrounding areas of bronchopneumonia and in the small peripheral bronchioles. The bronchioles at first were infiltrated with plasma cells and lymphocytes; the lumens were flattened and later entirely obliterated. The course of events is depicted diagrammatically. In a cavity with tuberculous bronchopneumonia, artificial pneumothorax may kink the draining bronchus which is diseased and structurally weakened. This may result in a tension cavity. This, in turn, causes alteration of the alignment of diseased bronchioles in the vicinity with early collapse and infection of the alveoli. This final picture is the so-called opaque lung or atelectasis.—*"Atelectasis" during Collapse Therapy for Pulmonary Tuberculosis, J. Cuthbert & M. N. Nagley, Tubercle, July, 1948, 29: 154.—(A. G. Cohen)*

Pulmonary Circulation.—Studies of cardiac output and pulmonary arterial pressure were performed using the venous catheter technique in 3 normal individuals and in 8 patients with various types of chronic pulmonary disease. Measurements were made at rest and during exercise on a stationary bicycle.

Two of the 3 normal subjects showed a decrease in the mean pressure in the pulmonary artery during exercise; all showed a marked drop in pulmonary vascular resistance and a minimal increase in the work of the right ventricle during exercise. Three of the patients with chronic pulmonary disease showed a significant elevation of pulmonary arterial pressure at rest, and in all 8 cases the mean pressure increased during exercise. There was either no change or an increase in the pulmonary vascular resistance during exercise, and the work of the right ventricle was invariably higher than in the normal subjects at a corresponding work level. The findings indicate that the expansibility of the pulmonary vascular bed during exercise is limited in patients with chronic pulmonary disease. Anoxia may contribute to the elevation of pulmonary arterial pressure during exercise in those patients whose arterial oxygen saturation falls. The physiological evidence upon which an estimate of disability must be based in patients with chronic pulmonary disease is extended by these studies of the pulmonary circulation during exercise. (Authors' summary.)—*Studies of the Pulmonary Circulation at Rest and during Exercise in Normal Individuals and in Patients with Chronic Pulmonary Disease, R. L. Riley, A. Himmelstein, H. L. Motley, H. M. Weiner & A. Cournand, Am. J. Physiol., February 1, 1948, 152: 372.—(G. C. Leiner)*

Pressure Breathing and Cardiac Output.—Three types of intermittent positive pressure breathing have been differentiated by the shape of the curve of the pressure in the mask and have been correlated with the changes in cardiac output observed in 33 experiments on 29 human subjects. The three types of pressure curves were as follows: type I, symmetrical with gradually increasing and decreasing slopes and expiratory and inspiratory times approximately the same, and the final expiratory pressure above the atmospheric level; type II, asymmetrical with the pressure rapidly increasing during inspiration and rapidly dropping during expiration, long inspiratory and short expiratory intervals, and the final expiratory pressure above the atmospheric level; and type III, asymmetrical with the pressure gradually increasing during inspiration and suddenly dropping early in expiration to the atmospheric level, the expiratory time equaling or exceeding the inspiratory. Cardiac output was decreased more or less in proportion to the increase in mean mask pressure with curves of the first and second types. There was no decrease in cardiac output with the third type of curve. The net filling pressure of the right ventricle decreased as the mask pressure rose and increased as the mask pressure fell with all types of curves. The mean net filling pressure of the right ventricle was calculated for complete respiratory cycles during intermittent pressure breathing and during ambient breathing. This mean pressure was reduced in cases where the cardiac output fell during intermittent pressure breathing. Conversely, when the cardiac output was increased, the mean net filling pressure rose. Interpreted in terms of variation in the stroke volume, these changes suggest that the deficit in cardiac output, which was incurred during the inspiratory phase, is compensated for under the expiratory phase. When the pressure drop is rapid in expiration so that the resulting intrapleural pressure is low and the right ventricular net filling pressure high, compensation is complete if the expiratory time is of sufficient duration. The expiratory time must equal or exceed the inspiratory time in order that the number of heart beats during expiration may equal or exceed the number during inspiration. With the third type of curve, the time and pressure relationships permit complete compensation and the mean right ventricular net filling pressure is not decreased. With the first and second types, compensation is incomplete because the mask pressure does not drop rapidly with the first type. The expiratory time is too short with the second. Intermittent positive pressure breathing should provide a mask pressure curve that shows: (*1*) a gradual increase in pressure during inspiration, (*2*) a subsequent rapid drop

in pressure, (3) a mean mask pressure during the expiratory period as near the atmospheric level as possible, and (4) an expiratory time equal to or exceeding the inspiratory time. Adequate ventilation can be provided with the above type of pressure breathing in man with a minimal disturbance to the circulation, and this type would seem most desirable physiologically for administering artificial respiration.—*Physiological Studies of the Effects of Intermittent Positive Pressure Breathing on Cardiac Output in Man, A. Cournand, H. L. Motley, L. Werko & D. W. Richards, Jr., Am. J. Physiol., January, 1948, 152: 162.*—(*G. C. Leiner*)

Alveolar Pressure.—Measurements of the alveolar pressure and the simultaneous rate of flow of respired air in 21 human subjects are presented. The data for each subject can be described by an equation of the form $P = K_1V + K_2V^2$, where P is the alveolar pressure and V is the volume rate of flow. The constants K_1 and K_2 differ for different individuals. In the average subject an alveolar pressure of 1.8 cm. of water is required to produce a flow of 500 cc. per second. (Authors' summary.)—*Measurement of Alveolar Pressure in Human Subjects, A. B. Otis & D. F. Proctor, Am. J. Physiol., January 1, 1948, 152: 106.*—(*G. C. Leiner*)

Myocardial Metastases from Bronchogenic Carcinoma.—The purpose of the article is to correlate the occurrence of arrhythmias and auricular involvement in an unselected series of 148 cases of bronchogenic carcinoma. Myocardial involvement was demonstrated in 17 cases. Of these, 11 showed arrhythmias, consisting of paroxysmal auricular fibrillation in 5, permanent auricular fibrillation in 2, permanent auricular flutter in 2, paroxysmal flutter and fibrillation in one and paroxysmal extrasystoles in one. There was no rheumatic history or postmortem evidence of rheumatic fever, hypertension or degenerative heart disease in any case. The average age was 54.7 years in cases with arrhythmia and 44.0 years in cases with regular rhythm. The primary lesion was in the left lung in 8 cases and in the right in 9. The left auricle was involved in 14 cases, the right auricle in 7, the left ventricle in 2 and the right ventricle in none. The histological type was squamous cell in 5 and oat-cell in 8; one each showed giant and spheroidal cell, oat and spheroidal cell and undifferentiated carcinoma. Serial sections of the coronary arteries were not made so that it cannot be stated for certain that arteriosclerosis was not a factor in producing the arrhythmia in some cases. It is assumed, however, that malignant invasion of the myocardium was at least the major factor. In 358 cases of nonbronchial carcinoma, metastases to the myocardium were found in 19 cases. The lesion was primary in many organs but malignant melanoma and some sarcomas showed a particularly high incidence. None arose from the gastro-intestinal tract below the esophagus. Only one showed an arrhythmia. Thus, the incidence of myocardial metastases from bronchogenic carcinoma is greater than that from all other sources combined.—*Myocardial Metastases from Bronchial Carcinoma and Other Neoplasms, J. E. G. Pearson, Brit. J. Tuberc., April, 1948, 42: 31.*—(*A. G. Cohen*)

Action of Diaphragm in Emphysema.—The range of the diaphragmatic movements was examined under costal and abdominal pressure in 9 normal and in 34 emphysematous subjects. Costal pressure enabled the normal, but not the emphysematous subject, to lower the diaphragm more on maximum inspiration. Abdominal pressure enabled both groups to push the diaphragm higher up in maximum expiration; during quiet breathing it restricted the diaphragmatic movements to a minimum. In no cases did improvement of the vital capacity occur. Under costal pressure the vital capacity was always considerably reduced. Its reduction under abdominal pressure was much smaller. Nevertheless, patients who used belts for several weeks were found to have an increased vital capacity after the belts were removed. This improvement is due to the additional respiratory training

which the compression produced.—*The Influence of Costal and Abdominal Pressure on the Action of the Diaphragm in Normal and Emphysematous Subjects, H. Herxheimer, Thorax, June, 1948, 3: 122.*—(*A. G. Cohen*)

Electrocardiogram in Pneumothorax.—Electrocardiographic studies were made before and after induction of a therapeutic pneumothorax. Ten patients with right pneumothorax and 13 patients with left pneumothorax were examined. P-wave changes were not consistent in left pneumothorax; in right pneumothorax P_1 showed a tendency towards decrease in amplitude, P_3 showed a tendency towards increase. P-R intervals remained unchanged. In right pneumothorax, there was a tendency toward decrease of QRS in lead 1. There was a tendency toward a shift of the axis to the right, more so in right pneumothorax than in left pneumothorax. T_1 became smaller in right and left pneumothorax. T_3 became larger in 10 of 13 cases of left pneumothorax; in right pneumothorax, T_3 showed no consistent change. In right pneumothorax, there were no important changes of the QRS complexes in the precordial leads. In all cases of left pneumothorax, the QRS complexes became smaller in two or more precordial leads; in 11 of the 13 cases the QRS complexes became more inverted in two or more precordial leads. In right pneumothorax there were no consistent precordial T-wave changes. In all cases of left pneumothorax the T-waves became smaller or inverted in two or more precordial leads. After resorption of the pneumothorax air the electrocardiographic changes disappeared. There was no correlation of the electrocardiographic changes with the degree of cardiac displacement or with the presence or absence of adhesions.—*Electrocardiographic Changes in Pulmonary Collapse Therapy. I. Artificial Pneumothorax, D. Feldman & C. Silverberg, Am. Heart J., May, 1948, 35: 800.*—(*G. C. Leiner*)

Venous Pressure in Tuberculosis.—Abnormal venous pressures have been observed frequently in cases of thoracoplasty, intrapleural and extrapleural pneumothorax. Measurements of the peripheral venous pressure of 55 patients with pulmonary tuberculosis confirm earlier reports that pressure changes are inconstant, often transient, and usually present only on the side of the more extensive disease or of the collapse. Among the patients were 30 who were dyspneic at rest but in whom clinical and roentgenographic evidence of right heart failure was absent or questionable. In such patients, venous pressure determinations offer valuable help in detecting early right heart failure. Collapse therapy does not interfere because it produces a unilateral increase in the venous pressure. The presence of the hepatojugular reflux phenomenon of Pasteur and Rondot is further evidence of heart failure; it is not produced by collapse therapy.—*The Relationship of Peripheral Venous Pressures to Pulmonary Tuberculosis, A. Paley & M. M. Alexander, A. J. M. Sc., February, 1948, 215: 189.*—(*G. F. Mitchell*)

Pneumopericardium from Pneumothorax.—In 2 cases receiving artificial pneumothorax for pulmonary tuberculosis, pneumopericardium appeared. The pneumothorax was on the right in one and on the left in the other. The pneumopericardium rapidly absorbed following abandonment of the pneumothorax. It is felt that the underlying mechanism is a congenital pleuropericardial defect.—*Pneumopericardium Complicating Pneumothorax Therapy, P. Ellman & K. K. Hussain, Thorax, September, 1948, 3: 129.*—(*A. G. Cohen*)

Prognosis of Cavitary Tuberculosis.—Positive sputum is regarded as the criterion of the presence of a cavity. The fate, after 20 years, of 1,233 unselected patients with positive sputum was investigated. There was found to have been improvement in the number of patients surviving three and five years from 1934, 1935 and 1936 onward. There was also improvement in the eight year survival period during 1937 to 1939. This

improvement was due to the various advances in treatment and care that have taken place. There was insignificant improvement in the ten year survival period. These studies indicate that the greatest care must be used in evaluating the apparent efficacy of any new type of treatment.—*The Fate of the Tuberculous Cavity, B. Mann, Tubercle, June, 1948, 29: 181.*—(*A. G. Cohen*)

Therapeutic Pneumoperitoneum.—Seventy-four patients with pulmonary tuberculosis were treated by phrenic crush supplemented by pneumoperitoneum. The treatment was used in cases falling into several categories. As a supplement to a partially effective pneumothorax, it was successful in 3 and partly successful in one of 8 cases. The good results were obtained chiefly with basal or mid-zonal cavities. As the sole definitive measure, it was successful in 14 and partly successful in 4 of 35 cases. The good results were obtained chiefly with exudative disease, infiltrations without cavitation, and cavities with no significant fibrosis. As a preparation for major surgical procedures, both on the homolateral and contralateral sides, it was effective in accomplishing the limited objective in 16 of 19 and 5 of 8 cases, respectively. It failed in all 4 cases of far advanced disease. The chief complications were effusion in 4 cases, including one of fatal peritonitis, and a non-fatal air embolism in one case. The patients experienced very little discomfort. The pneumoperitoneum was maintained as long as pneumothorax would have been in the same case.—*Pneumoperitoneum: Its Place in Treatment, R. J. Keers, Brit. J. Tuberc., July, 1948, 42: 58.*—(*A. G. Cohen*)

Absorption of Pneumoperitoneum.—The rate of absorption of pneumoperitoneum was studied in 33 cases of pulmonary tuberculosis. The pneumoperitonea were classified arbitrarily as full, moderate and shallow. The average time required for absorption was eight weeks in the full, five and a half weeks in the moderate and two and a half weeks in the shallow. The rate of absorption of air was not affected by exercise, duration of the pneumoperitoneum or the presence of fluid. Ex-vacuo effusions did not develop.—*The Absorption of Pneumoperitoneum, W. Fox, Thorax, September, 1948, 3: 141.*—(*A. G. Cohen*)

Extrapleural Pneumothorax.—Extrapleural pneumothorax was employed in 128 patients with pulmonary tuberculosis. All but 5 patients were followed for at least five years or until death. Fifty-six (44 per cent) were dead, 58 (45 per cent) were alive and well while 14 (11 per cent) were alive but with symptoms. In 28 cases the extrapleural pneumothorax was used with an intrapleural pneumothorax on the same side ("combined" operation). In the other 100 cases, the extrapleural operation alone was done ("standard" operation). The mortality was 14 per cent in the "standard" and 21 per cent in the "combined" group. Tuberculous infection of the extrapleural space was detected in 13 per cent of the "standard" and 28 per cent of the "combined" group. Other frequent complications were nontuberculous infection of the space, bronchopleural fistula, hemorrhage. atelectasis and effusion. Very few late complications were seen in cases where a satisfactory pneumothorax had been obtained. In 27 of the 33 cases in which the pneumothorax was voluntarily abandoned after an average period of five years, there were no relapses or complications. Factors leading to a successful outcome of the operation were (*1*) a short period of symptoms, (*2*) a small cavity which was not obviously subpleural, and (*3*) minimal disease in the contralateral lung.—*Extrapleural Pneumothorax, A. T. M. Roberts, Thorax, September, 1948, 3: 166.*—(*A. G. Cohen*)

Open Intrapleural Pneumonolysis—In 13 cases of pulmonary tuberculosis receiving pneumothorax, adhesions were severed by an open operation. In each case, a closed pneumonolysis had been attempted previously. The advantages of salvaging an ineffective pneumothorax are obvious. The disadvan-

tages of this procedure are: (1) the operator may not be able to cut all the adhesions, and (2) the operation carries with it all the complications of pneumothorax, thoracoscopy and thoracotomy combined. The chief complications are pleural infection and breakdown of the suture line. Early results in this series were excellent improvement in 7 cases, good progress in 2 and no change in one. One patient is worse and 2 have died; in these 3, there was a complicating empyema.—*Open Intrapleural Pneumonolysis in the Treatment of Pulmonary Tuberculosis, R. A. Smith, Brit. J. Tuberc., July, 1948, 42: 51.*—(*A. G. Cohen*)

Tuberculous Lesions of the Spine.—Tuberculosis of the spine is primarily a destructive disease with little signs of bone repair in the early stages but it may be sclerotic. The intervertebral space is often preserved in spite of extensive destruction of several vertebral bodies. Later, narrowing of the intervertebral space may result from extrusion of the nucleus pulposus into the softened bony or ligamentous tissue. In contrast, other infections often cause early and complete destruction of the vertebral discs. Infections limited to the neural arches are practically always nontuberculous. The spread in tuberculosis occurs by extension under the anterior longitudinal ligaments with surface invasion of the vertebral bodies anteriorly. In contrast, other infections spread by direct extension from vertebral body to adjacent body through the intervertebral disc. The classical description of bone tuberculosis is that of bone destruction without sclerosis and with minimal new bone formation. During recent years, however, reports of tuberculosis of the spine have emphasized sclerosis as a conspicuous feature, even in the absence of secondary infection. The lesions may be very extensive and in one case there were tuberculous lesions in all stages of development in at least 8 vertebral bodies.—*Multiple Tuberculous Lesions Of The Spine, J. L. Feuchtwanger, Brit. J. Radiol., August, 1948, 21: 400.*—(*B. Hyde*)

New Mycobacterial Infection in Man.—The author reports 6 patients with cutaneous ulcers in which strongly acid-fast bacilli were demonstrated by biopsy. The clinical, histological, and bacteriological characteristics of all the cases were similar. The lesions were solitary and initially small and single; later, further ulceration occurred near the original site. All were found on the extremities. Each lesion appeared as an inconspicuous, solitary area of slightly inflamed induration in which a small ulceration appeared and then extended indolently. The ulcer was refractile to therapeutic measures which were followed by edema, pyogenic reactions, and often acceleration of the destructive process. Sloughing and denudation of large areas were not accompanied by systemic reaction unless complications occurred. The lesion spread by extension of the marginal induration which subsequently broke down, exposing perpendicular or undermined edges of necrotic tissue. A striking feature was the development from the exposed fascia of a gelatinous mass, "like blubbery granulation tissue." Histological examination of tissue from the ulcer showed necrosis of the ulcer wall, the necrosis being most extensive in fatty tissue in which it extended beneath the dermis. Acid-fast bacilli were found in enormous numbers, in every instance grouped characteristically in sharply defined oval or round masses often as great in diameter as fat cells. Others of the bacilli were in smaller, more scattered groups. No tubercular follicles, giant cells, endothelioid grouping or caseation were seen. Pus from a patient's lesion was treated with sulphuric acid and injected subcutaneously into rats, producing ulceration of the epididymis. Rabbits and mice inoculated intraperitoneally developed similar but less extensive lesions. Acid-fast bacilli were demonstrated in the ulcers of all the animals. Guinea pigs inoculated by either route failed to develop progressive disease. The acid-fast organisms were cultured on such solid media as egg yolk agar, blood agar and the media of Petragnani, Loeffler, and Dorset. The optimum temperature for growth was 30

to 33°C.—*A New Mycobacterial Infection in Man: I. Clinical Aspects, P. MacCallum; II. Experimental Investigations in Laboratory Animals, J. C. Tolhurst & G. Buckle; III. Pathology of the Experimental Lesions in the Rat, H. A. Sissons; IV. Cultivation of the New Mycobacterium, G. Buckle & J. C. Tolhurst, J. Path. & Bact., January, 1948, 60: 93.*—(*H. J. Henderson*)

Spontaneous Pneumothorax.—In 1943 the United States Army had 873 hospital admissions for simple benign spontaneous pneumothorax. It has become apparent in recent years that this type of pneumothorax is relatively common, especially in healthy young adult males. Pleural fluid is slight, fever is uncommon, and uncomplicated re-expansion is the rule. Sixty-three cases are reported in detail. Half of the patients were between 20 and 30 years of age; the ratio of males to females was 5 to 1. In 70 per cent the time required for re-expansion was seven weeks or less but, in any particular patient, it was difficult to predict the time which would be required. In only 3 per cent physical exertion preceded the onset of the pneumothorax. Chest pain occurred in all and subsided in a few days. Dyspnea was present in 83 per cent; 5 patients were slightly cyanotic at the onset. Four patients had no pleural fluid; two had fluid above the diaphragm and in both cases gross blood was aspirated. No patient had pleural adhesions demonstrable on chest X-ray, in sharp contrast to patients with active pulmonary tuberculosis. The sedimentation rate was normal in 73 per cent and slightly elevated in 27 per cent of the cases. Of those who had white blood counts on admission, 70 per cent were normal and the others showed only slight elevation. Most patients were afebrile but two had fever for more than one week. They both had thrombophlebitis. Recurrences occurred in 19 per cent, usually on the same side, but the prognosis was excellent.—*Benign Idiopathic Spontaneous Pneumothorax, A Review of Sixty-three Cases.—B. Hyde & L. Hyde, Am. J. M. Sc., April, 1948, 215: 427.*—(*G. F. Mitchell*)

Spontaneous Mediastinal Emphysema.—A 23 year old man developed pain in the left chest which radiated to the left shoulder and left arm. He became dizzy, nauseated and went into shock. Auscultation revealed a crunching sound over the heart during systole and diastole. The diagnosis of acute myocardial infarction was made. A chest roentgenogram (posteroanterior view) taken a few days later showed no abnormality. A few weeks later the patient developed similar signs and symptoms. A lateral chest roentgenogram revealed an area of increased translucency anterior to the heart. The diagnosis of recurrent spontaneous mediastinal emphysema was made.—*Recurrent Spontaneous Mediastinal Emphysema Simulating Myocardial Infarction, P. C. Pellegrino & E. N. Silber, Am. Heart J., September, 1948, 36: 447.*—(*G. C. Leiner*)

Suppurative Bronchopneumonia. — The authors saw approximately 3,500 cases of pneumonia at a naval hospital during the winter and spring of 1944–1945. Seven cases of bronchopneumonia were characterized by parenchymal suppuration and necrosis with cavity formation and are reported in some detail. Other investigators have reported an equal incidence in the sexes and an occurrence in all age groups, but there appears to be a preponderant frequency in children and young adults. In 4 of the authors' cases, a pure culture of beta hemolytic streptococcus was found; in the other 3 no pathogenic organisms were identified. The clinical symptoms of suppurative bronchopneumonia are fever, cough and expectoration of purulent sputum, with a frequent history of antecedent upper respiratory tract infection. Pleuritic pain is common. There are 4 clinical types of suppurative bronchopneumonia: (*1*) An ordinary bronchopneumonia except for the roentgenographic demonstration of a cavity. Recovery is usually uneventful. (*2*) A more severe, protracted disease which tends to

spread to new bronchopulmonary segments. Some patients recover spontaneously after weeks or months of prolonged relapsing illness. In others, chronic bronchiectasis develops secondary to the pulmonary suppuration and the superimposed atelectasis with its resultant interstitial fibrosis. (*3*) Bronchopneumonia with local surgical complications such as aerobic pulmonary abscess or pleural involvement with empyema or pyopneumothorax. (*4*) Bronchopneumonia in which general or regional spread occurs with cerebral involvement, diffuse septic embolic phenomena, mediastinitis or pericarditis. The cardinal feature of the roentgenographic findings is the presence of one or more areas of cavitation within an area of pneumonic consolidation. The early appearance of a cavity with a sharply defined border and an air-fluid level usually makes differentiation possible. The diagnosis of suppurative bronchopneumonia usually is made without difficulty from the clinical and roentgen findings. However, the more severe forms must be distinguished from other diseases such as pulmonary tuberculosis, putrid lung abscess, bronchogenic carcinoma, actinomycosis and primary atypical pneumonia. Pulmonary abscess incidental to extensive necrosuppurative bronchopneumonia and pulmonary abscess as the predominant feature with a surrounding necrosuppurative pneumonic infiltration require no surgical intervention. The disease usually resolves spontaneously; less frequently it eventuates in bronchiectasis. The pulmonary and pleural complications, however, may require surgical management. The use of penicillin has resulted in a marked reduction in toxicity and may have prevented the occurrence of the more serious complications. — *Suppurative Bronchopneumonia with Cavitation, S. P. Perry & R. Shapiro, Radiology, March, 1948, 50: 851.—(G. F. Mitchell)*

Acute Benign Dry Pleurisy.—An outbreak is described in which 5 nurses and one ward maid at a hospital were affected. All the cases appeared within six days of each other. The affected persons worked in scattered parts of the hospital. The duration of the disease was seven to fourteen days with an average of nine. Chest pain was present in every case and signs of dry pleurisy in 3. Fever ranged up to 103°F and lasted about five days. The elevation in pulse rate was proportional to the fever. In one of the cases there was an area of hyperaesthesia on the chest wall. Roentgenograms were normal. Two cases showed a neutropenia. Sedimentation rates were normal.—*Acute Benign Dry Pleurisy, J. D. Gray & F. S. Carter, Lancet, July 31, 1948, 2: 254.—(A. G. Cohen)*

Calcium Gluconate for Pleuritic Pain.—Clinical observations have indicated that the origin of pleuritic pain is more complex than simple friction between the pleural surfaces. The relief of acute pleural pain by the intravenous injection of calcium gluconate is reported. Ten to 20 ml. of a 10 per cent solution were given in two to four minutes. All but 3 of 30 patients experienced definite relief, the effect being evident in about sixty seconds. Any residual pain was described as a dull aching and unrelated to breathing. Two of the 3 patients obtaining no relief were not benefited by procaine block of the intercostal nerves. Only 4 patients were completely relieved and there was a gradual return of pain in thirty to sixty minutes. Tenderness and hyperasthesia of the chest wall or upper abdominal muscles also disappeared to reappear with the return of pleuritic pain. Ten patients given a second injection were again relieved. The practical value of the treatment lies in its simplicity and its usefulness when examination of the patient is difficult or when he cannot cough to obtain sputum for examination. In 1940 Buchtal reported electromyographic studies of patients with painful muscles and demonstrated that various drugs, including calcium salts, brought about the relaxation of muscle spasm. Other reports emphasize that calcium injection aids in the restoration of normal muscle tone. Several investigators have demonstrated the relief of pain by the local injection of procaine, one injection often

giving permanent relief. Kelly was able to relieve pain in both acute pleuritis and fibromyositis in this manner. Dybdahl relieved pleuritic pain by spraying ethyl chloride on the skin over the painful area. The authors report success with this method also, the spray being used for twenty to thirty seconds. These results may be explained by the so-called "chain" effect of muscle spasm described by Payr. Pain originating anywhere in the "chain" leads to painful muscle spasm and the pain of spasm induces still more spasm and pain. Thus, if the chain is broken at any point, the spasm is relieved. The relief of painful muscle spasm by the use of erythriodine in anterior poliomyelitis and the relief of painful muscle spasm by curare has been repeatedly demonstrated. Thus, much of the pain of pleuritis may be due to painful spasm of the intercostal muscles which perpetuates itself in a vicious cycle of spasm, pain and more spasm. Relief by any method seems to depend upon interruption of the cycle at some point.—*Pleuritic Pain: Use of Intravenous Calcium Gluconate in Its Relief, I. L. Bennett & W. Lathem, Am. J. M. Sc., April, 1948, 215: 431.*—(*G. F. Mitchell*)

Boeck's Sarcoid.—This report is derived from the study of 7 cases of Boeck's Sarcoid, including one death and autopsy. All the patients were young adults and 4 were Negroes. Five patients had widened hilar shadows on roentgenograms. Tuberculin tests were done on 6 patients and were negative in all. The sedimentation rate was elevated in 5 and remained so for prolonged periods; this test may measure the activity of the disease. In the 2 patients on whom serum protein determinations were carried out, the globulin fraction was above normal. Final diagnosis depends upon histological study of a biopsy specimen, usually an enlarged peripheral lymph node. Several biopsies may be necessary. In one patient, the diagnosis was established by biopsy of a nodule in the biceps muscle. One patient died suddenly at a time when he appeared to be improving and was symptom free. The striking postmortem finding in this man was extensive sarcoid infiltration of the myocardium. He had maintained a pulse rate of 90 to 120 even when afebrile. In retrospect, the authors feel that persistent tachycardia should arouse suspicion of myocardial involvement in Boeck's sarcoid. One other patient with persistent tachycardia manifested electrocardiographic findings indicative of myocardial damage.—*Boeck's Sarcoid: Observations of Seven Patients, One Autopsy, G. S. Bates & J. M. Walsh, Ann. Int. Med., August, 1948, 29: 306.*—(*H. R. Nayer*)

Histoplasmosis and Torulosis of Adrenals.—Tuberculosis has long been recognized as the most frequent infectious cause of adrenal insufficiency though other organisms may produce the same signs and symptoms. Despite their relative infrequency, fungus diseases, especially histoplasmosis and torulosis, should be kept in mind in the differential diagnosis. All the available cases of adrenal involvement by histoplasmosis and torulosis are reviewed. In all, 19 cases of involvement due to Histoplasma capsulatum have been reported. Some symptoms of adrenal insufficiency are reported but for the most part they are masked by complaints referable to other organs. Autopsy showed marked enlargement and parenchymal replacement by yellowish-gray fibrocaseous tissue containing the causative organism. Only 8 cases of adrenal insufficiency due to Torula histolytica have been reported. Two new cases, one of histoplasmosis and one of torulosis are reported by the authors. The symptoms of adrenal insufficiency were recognized in one of the cases prior to death. As in tuberculosis, the ratio of cases of adrenal failure due to histoplasmosis is 2 males to 1 female; 76 per cent of the histoplasmosis cases and only 28 per cent of adrenal torulosis cases showed caseation. The authors suggest that the caseous reaction may be an allergic one.—*Histoplasmosis and Torulosis as Causes of Adrenal Insufficiency, A. J. Rawson, L. H. Collins, & J. L. Grant, Am. J. Med., April, 1948, 215: 368.*—(*G. F. Mitchell*)

Arterio-venous Aneurysm of the Lung.—Two cases of arterio-venous aneurysm of the lung are reported, making a total of 25 cases on record. The outstanding feature is cyanosis. Dyspnea appears later and is progressive. It is probably due to stimulation of the respiratory center by the high carbon dioxide content of the arterial blood and not to cardiac failure. Cerebral anoxemia is responsible for the transient attacks of vertigo, faintness or convulsions which may occur. Pulmonary hemorrhage is frequent; capillary hemangiomata of the skin and mucous membranes are seen often. Clubbing of the fingers is common; hypertrophic osteoarthropathy is rare. There are no abnormal cardiac findings; blood pressure readings are normal. A murmur over the tumor is heard in about half of the cases. Secondary polycythemia and greatly diminished oxygen saturation of the arterial blood is characteristic. An opaque shadow representing the tumor is seen in the lung and may be connected to the hilus by vessels. It may decrease in size with Valsalva's experiment or increase in size when the patient attempts forced inspiration with mouth and nose closed. The lesion is a developmental malformation. The treatment is surgical removal by local excision, pneumonectomy or lobectomy. — *Arterio-venous Aneurysm of the Lung, C. G. Barnes, L. Fatti & D. M. Pryce, Thorax, September, 1948, 3: 148.*—(*A. G. Cohen*)

Lobar Adenocarcinoma.—The autopsy findings in 2 cases with extensive pneumonic consolidations are presented. In a 31 year old woman, diffuse adenocarcinoma of the right lung, with metastases to the left lung, hilar lymph nodes, adrenal gland and kidney was found. In a 73 year old woman the autopsy revealed diffuse adenocarcinoma of the right lung, with metastases to the left lung, hilar lymph nodes, adrenal glands and liver. No distinct point of origin of the tumors was found. The following conclusions are made: "(*1*) The growths are primary in the lung, but metastases from a latent primary glandular focus outside the lung should be carefully excluded. (*2*) They arise from the mucosa of the bronchial tree beyond the larger bronchi, the point of origin being obscured by the extensive growth involvement. (*3*) A source of origin from pre-existing epithelized alveoli cannot be proved. (*4*) A multicentric origin cannot be established. (*5*) To date there is no proved relationship between diffuse adenocarcinoma of the lung and pulmonary adenomatosis."—*Lobar Adenocarcinoma of the Lung Simulating Pneumonia. Report of Two Cases, G. Silverman & A. Angrist, Arch. Int. Med., March, 1948, 81: 369.*—(*G. C. Leiner*)

Horner's Syndrome from Osteochondroma.—Horner's Syndrome may be produced by any type of apical lung tumor but is most commonly associated with malignant neoplasms. The author reports the case history of a 34 year old woman in whom the syndrome had been present for twenty years. Intermittent gnawing pain in the left arm had been present since the age of seven. Chest X-ray revealed a left apical mass with irregular calcification and a clearly demarcated outer margin. A hard bony mass was palpable in the left supraclavicular fossa. At operation the tumor mass was found to arise from the first rib and was completely removed. The pathological diagnosis was osteochondroma. Eight months after removal, there was evidence of some regeneration of the sympathetic fibers. Horner's Syndrome produced by osteochondroma of left first rib is an extremely rare finding.—*Horner's Syndrome Due to An Osteochondroma of the First Rib, J. F. Simpson, Canad. M. A. J., August, 1948, 59: 152.*—(*H. R. Nayer*)

Roentgenography of the Diaphragm.—The diaphragm, both anatomically and physiologically, is a vulnerable structure, being readily involved by pathological changes arising from above and below or from inherent weaknesses of the structure itself. Roentgenographic studies have paid the greatest attention to the anteroposterior projection, although the surface of the diaphragm is not horizontal throughout. Each leaf has the

shape of a dome, the highest point being medial and anterior with a downward slope in the lateral and backward extensions, so that only the surface of the highest level can be seen in an anteroposterior view. One-third to one-half of the diaphragm lies behind the heart and is not seen at all. Thus, for a satisfactory roentgenographic examination of the entire diaphragm, a lateral as well as a frontal view is essential. This technique has been used successfully for many years and the authors have demonstrated many abnormal conditions which were hardly suspected in the customary frontal projections. The diaphragm, being in intimate contact with the underlying structures, cannot be seen by itself, but the presence of free air within the abdomen enables one to see it elevated, giving a more accurate picture of its position, contour, and thickness. A study of 8 cases showed that the fundamental roentgenographic technique of making two projections at right angles to one another should be applied to the diaphragm to demonstrate satisfactorily its complete topography. It has made possible the recognition and localization (with or without the aid of barium suspension) of herniation through any part of the diaphragm; the differentiation of lesions not seen in the anterior projection; demonstration of elevation of the diaphragm, in whole or in part, often suggestive of intra-abdominal tumors; and finally, the most practical application of all, the diagnosis of subphrenic abscess. This is indicated by elevation of the diaphragm and obliteration of the posterior costophrenic angle producing the "plateau sign of subphrenic abscess."—*The Diaphragm, S. Brown & A. Fine, Radiology, February, 1948, 50: 157.* —(*G. F. Mitchell*)

Pleural Fluid Simulating Elevated Diaphragm.—The author presents 5 cases in which pleural fluid at the lung base closely simulated elevation of the diaphragm, in 2 instances on the right and in 3 on the left. The roentgenographic appearance of the fluid must be distinguished from subphrenic disease (inflammatory or neoplastic), paralysis of the diaphragm, atelectasis, eventration of the diaphragm, and intrapleural or intrapulmonary neoplastic disease. It is most difficult to differentiate between the collection of fluid and a subphrenic disease producing a progressive elevation of the diaphragm. The patients often have fever and vague symptoms which are of little help in localizing the process. The signs on physical examination may be identical in the two conditions. Fluoroscopy, lateral decubitus roentgenograms, pleural tap, demonstration of the stomach bubble, which may be accentuated by giving the patient a carbonated drink prior to examination, and pneumoperitoneum are suggested as useful procedures in differential diagnosis. Among the factors leading to the production of basal fluid accumulations, fibrinoplastic adherence of visceral and parietal layers of the pleura is believed to be of major importance.—*Basal Pleural Fluid Accumulations Resembling Elevated Diaphragms, D. B. Jones, Radiology, February, 1948, 50: 227.*—(*G. F. Mitchell*)

Pleural Fluid Simulating Elevated Diaphragm.—The authors present a typical example of an infrapulmonary effusion masquerading as an elevated diaphragm. A 27 year old white female with bilateral pulmonary tuberculosis complained of pain in the right lower portion of the chest and became slightly febrile. Fluoroscopy revealed an elevated, arched right diaphragmatic leaf which moved over only one interspace. The costophrenic angle was clear and no free fluid was detected. Roentgenograms confirmed these findings. The lower half of the right side of the chest was dull to percussion, with decreased or absent breath sounds. Diagnostic pneumoperitoneum was initiated and demonstrated an infrapulmonary effusion. An anteroposterior roentgenogram of the chest in the right decubitus position revealed a well defined fluid level in the costal gutter, some of the fluid remaining in the infrapulmonary space at the end of five minutes. Diagnostic thoracentesis revealed a clear amber fluid which contained no acid-fast bacilli.—*Infrapulmo-*

nary Effusion Masquerading as an Elevated Diaphragm, J. T. Taguchi & S. Dressler, Radiology, February, 1948, 50: 223.—(G. F. Mitchell)

Diagnosis of Retrocardiac Shadows.—The configuration of the cardiac shadow observed on a routine roentgenogram of the chest was given adequate emphasis and the findings were correlated with electrocardiograms by Master; the variations in density within the triangular shadow have received little consideration unless there was a fluid level or calcification. Systematic analysis of the triangular shadow will frequently disclose considerable variations in density, many of which present characteristic features of diagnostic importance, leading to other diagnostic procedures or aiding in the detection of differential diagnosis and of asymptomatic retrocardiac lesions. The space-occupying lesions located in the upper half of the mediastinum produce pressure signs relatively early and are frequently suspected by the clinician. In the retrocardiac region, where the space is wide, the most obscured thoracic lesions occur and these may reach considerable size before they lead to clinical signs or symptoms. By early detection and proper evaluation of the abnormal retrocardiac shadows, the radiologist may establish the diagnosis before clinical signs and symptoms become apparent. Certain diseases of the stomach, esophagus, aorta, heart, lungs, and vertebral columns cast characteristic retrocardiac shadows which may not project beyond the cardiac outline, but which can be recognized as variations in density within the cardiac shadow. Multiple convex lines within the cardiac shadow indicate the presence of the stomach in the retrocardiac region. A simple double contrast method for the study of the cardiac-esophageal junction and the relationship of the lower third of the esophagus to the stomach is presented.—*Differential Diagnosis of Retrocardiac Shadows, S. S. Nemec, Radiology, February, 1948, 50: 174.—(G. F. Mitchell)*

Visible Bronchial Tree.—The visibility of the air-filled bronchi within parenchymatous consolidations of the lung is explained on theoretical grounds and illustrated by the use of a model made of a box containing two rows of drinking straws; one row, empty, *i.e.*, air filled, and the other filled with paraffin. A roentgenogram of the box showed the paraffin-filled straws as white bands; the empty straws are invisible except for the faint shadows cast by their walls. In a roentgenogram of the box filled with water, the paraffin-filled straws were not detectible; the air-containing straws appeared as dark bands. The "visible bronchial tree" is a helpful sign in the recognition of parenchymatous lesions of the lung, especially of pneumonic consolidation, including tuberculous pneumonia. Infarcts and tumors may also show this sign if the bronchi are not blocked or filled by blood or secretion. In atelectasis due to compression or obstruction, the approximation of the visible bronchi may reveal the atelectatic nature of the process. The "visible bronchial tree" is also helpful in differentiating basal pneumonic consolidations from pleural effusion or thickened pleura. Only the presence of the visible bronchial tree is of diagnostic value; it may be absent in pneumonic consolidation. Failure to demonstrate the bronchi may be due to filling with secretion, edema, or blood, or may be the result of technical errors, such as over or under exposure. The sign is particularly helpful in recognizing faint congestion and faint shadows in pulmonary consolidations. This sign has been found most valuable in pediatric roentgenology.—*The Visible Bronchial Tree, F. G. Fleischner, Radiology, February, 1948, 50: 184.—(G. F. Mitchell)*

Streptomycin-requiring *Mycobacterium Ranae*.—Approximately 25 billion organisms of a streptomycin-resistant strain of *M. ranae* were plated on a deficient medium containing streptomycin. A variant was found which requires streptomycin since neither the parent strain nor the variant grew on the deficient medium alone. If such a variant

can also develop from pathogenic tubercle bacilli, its significance in the chemotherapy of tuberculosis is evident.—*A Variant of Mycobacterium Ranae Requiring Streptomycin for Growth, D. Yegian & V. Budd, J. Bact., April, 1948, 55: 459.*—(*F. G. Petrik*)

Effect of Fluoride on Mycobacteria.—Recently Davis and Dubos have shown that fluoride, in concentrations that do not affect the growth of *M. tuberculosis* H37RV, inhibits the action of lipase on "Tween 80." The present study deals with the effects of the fluoride ion on the metabolism of certain mycobacteria. Most of the experiments were done with a rapidly growing BCG strain of *M. tuberculosis* (ATC 8240). Similar effects were, however, obtained with *M. tuberculosis* (ATC 607) and the strain H37RV. The results showed that the oxygen uptake of washed suspensions of the BCG strain is greatly increased by sodium fluoride and that the amount of increase depends on the relative concentrations of fluoride and bacteria as well as upon the hydrogen ion concentration. The effect is greatest at pH 6.0, intermediate at pH 6.7, and least at pH 7.8.—*The Effect of Sodium Fluoride on the Metabolism of Certain Mycobacteria, R. J. Fitzgerald & F. Bernheim, J. Bact., May, 1948, 55: 677.*—(*F. G. Petrik.*)

Penicillin and Growth of Mycobacterium.—The effect of penicillin on the growth of *M. tuberculosis* was studied using four strains from active human cases in addition to standard laboratory strains of human, bovine and avian type. Crystalline penicillin G sodium (Merck) was used in a culture medium prepared according to the formula of Dubos and Davis. Human, bovine and avian types of *M. tuberculosis* were inhibited by concentrations of penicillin varying from 1 to more than 200 units per ml. depending on the size of the inoculum. Among the human strains, no marked difference in sensitivity was observed between a standard laboratory strain (H37RV) and strains recently isolated. The strains of bovine and avian type were more sensitive to penicillin than human ones.—*The Effect of Penicillin on the Growth of Mycobacterium Tuberculosis in Dubos Medium. M. Solotorovsky, E. J. Bugie & B. M. Frost, J. Bact., April, 1948, 55: 555.*—(*F. G. Petrik.*)

Desensitization and Tuberculin Cytotoxicity.—Tuberculin (OT) caused specific suppression of the initial migration of leucocytes from splenic explants of tuberculous guinea pigs. This suppression was markedly reduced by specific desensitization. This finding indicates that the cytotoxic effect of tuberculin on tissue explants of tuberculin-sensitive animals is the result of the interaction of a sessile antibody with specific antigen.—*Tuberculin Reaction. II. Cytotoxicity of Tuberculin for Splenic Explants of Desensitized Tuberculous Guinea Pigs, W. F. Kirchheimer & R. S. Weiser, Proc. Soc. Exper. Biol. & Med., June, 1948, 68: 407.*—(*F. B. Seibert*)

Synthetic Phthioic Acid.—Certain synthetic fatty acid produced necrosis, abscess formation and foam cell nodules in the omentum, lymph glands and spleen when injected into the peritoneal cavity of the guinea pig. Naturally occurring phthioic acid produced the same reaction.—*The Pathogenic Effect of Phthioic Acid and its Synthetic Analogues, J. Unger, C. E. Coulthard & L. Dickinson, Brit. J. Exper. Path., August, 1948, 29: 322.*—(*H. J. Henderson*)

Enzyme Inhibition by Streptomycin.—Benzoic acid is oxidized by an adaptive enzyme in certain *Mycobacteria*. Streptomycin inhibits this oxidation probably by inhibition of the formation of this enzyme. *Mycobacterium lacticola*, which oxidizes p-hydroxybenzoic as well as benzoic acid itself, was shown to have its ability to oxidize p-hydroxybenzoic acid less inhibited by streptomycin, if it had previously been grown in a medium containing 60 mg. per cent of p-hydroxybenzoate. Growth in benzoic acid increases to a small extent the amount of adaptive enzyme for p-hydroxybenzoic acid and growth in this latter acid has

a somewhat greater effect on the amount of adaptive enzyme for benzoic acid. It is clear that the two acids are oxidized by separate enzymes. Experiments were made with *Mycobacterium tuberculosis* BCG to determine the amount of benzoic acid required by the cell suspension to produce a measurable amount of adaptive enzyme and the time of exposure necessary for the effect to occur. Maximum inhibition by streptomycin occurs if it is added to the bacilli ninety minutes before the benzoate. The authors, however, have not been able so far to demonstrate adaptive enzyme formation in virulent *Mycobacteria* which are sensitive to streptomycin.—*The Inhibition by Streptomycin of Adaptive Enzyme Formation in Mycobacteria, R. J. Fitzergerald, F. Bernheim, & D. B. Fitzgerald, J. Biol. Chem., August, 1948, 175: 195.*—(*F. B. Seibert*)

Chemical Determination of Streptomycin.—A fluorometric method, based on the formation of a fluorescent hydrazone, for the determination of streptomycin in urine and tissue was described. The lower limit of sensitivity was 2 γ per ml. of urine and of tissue. Data on the analytical recoveries of streptomycin added to urine and to lung, liver, brain, heart, and spleen are presented. The utility of the method for the determination of streptomycin in urine and tissues following parenteral administration has been demonstrated.—*A Chemical Determination of Streptomycin in Body Tissues and Urine. V. C. Jelinek & E. Boxer, J. Biol. Chem., August, 1948, 175: 367.*—(*F. B. Seibert*)

Effect of Streptomycin and PAS on Tubercle Bacilli.—The growth of tubercle bacilli (H37RV) on Dubos medium is inhibited by 0.74 γ per cc. of streptomycin or by 1.2 γ of para-aminosalicylic acid (PAS). A moderately inhibiting concentration of either streptomycin or of PAS when combined with a completely noninhibiting concentration of the other drug produces a marked enhancement of inhibition of the growth *in vitro* of H37RV. *In vitro* growth of a resistant strain of human tubercle bacilli (H37RVNR1) is inhibited by 1.2 γ per cc. of PAS, and this inhibition is not enhanced by the addition of streptomycin.—*In vitro Effect of Streptomycin and Para-Aminosalicylic Acid (PAS) on the Growth of Tubercle Bacilli, K. Vennesland, R. H. Ebert & R. G. Bloch, Proc. Soc. Exper. Biol. & Med., June, 1948, 68: 250.*—(*F. B. Seibert*)

Streptomycin for Bone Tuberculosis.—Sixteen patients with tuberculosis of the bones and joints have been treated with streptomycin. Nineteen joints were involved. Sinuses were present in 4 patients. The period of observation following discontinuation of streptomycin therapy varied from one to eighteen months. In addition to streptomycin therapy, other forms of treatment were carried out whenever indicated. Response to streptomycin was favorable in 9 patients and fair in one patient. Four patients showed no benefit while treatment was too recent to permit valid conclusions in 2 patients. The daily administration of 1 Gm. of streptomycin for ninety days is considered a satisfactory course of treatment.—*Streptomycin in Tuberculosis of Bone and Joint, W. H. Bickel, H. H. Young, K. H. Pfuetze & T. Norley, J.A.M.A., June 19, 1948, 137: 682.*—(*H. Abeles*)

Streptomycin for Pulmonary Tuberculosis.—The authors outline some tentative clinical interpretations with regard to the use of streptomycin in pulmonary tuberculosis. Conclusions are based on their own experience and on direct knowledge of the work of others. In acute febrile tuberculosis, symptomatic response is often striking and may occur within three to four days, much before any changes in the specific lesions can be noted. Local symptoms, such as the pain of tuberculous laryngitis, cough or urinary symptoms may subside rapidly. Acute tuberculous lesions in the lung and larynx may show accelerated resolution under the influence of the antibiotic which suppresses bacterial growth before caseous necrosis has set in. Although complete clearing roentgenographically may occur, the likeli-

hood of persistent minute caseous foci must be assumed. Resolution of acute pulmonary lesions may proceed for several months after completion of a course of streptomycin. The presence of caseation limits, to a large extent, the response to streptomycin. In extensive caseous lesions, there may be early symptomatic response but little objective evidence of resolution and early relapse is common. The frequent finding of a caseous cerebral focus in tuberculous meningitis probably explains the high incidence of relapse and the fatality rate (approximately 80 per cent) after initial improvement. Definitive arrest of the tuberculosis still depends primarily on the slow healing and fibrosis of caseous disease. In spite of immediate favorable effects, streptomycin has, therefore, not shortened the time required for ultimate healing. Streptomycin may aid in restoring a favorable balance but the natural resistance of the patient remains of great importance. The greatest benefit from the drug may be obtained when it is employed in association with rigid rest. The partial improvement obtained with streptomycin may render surgical measures possible in previously hopeless cases. Drug-resistant bacilli appear within four weeks and are found in 70 per cent or more of patients within three to four months. Resistance appears to be more directly related to the duration of treatment than to the daily dosage. The authors' experience indicates that treatment for a period of six weeks with doses of 1 to 2 Gm. daily may be as effective as longer periods. Under the six week regimen, drug-resistance appeared in 25 to 30 per cent of the patients; the drug is, therefore, effective in the majority of these patients if further treatment is needed. In 12 of 18 patients, the authors observed complete loss of labyrinthine function when a dosage of 2 Gm. a day for six weeks was employed. Administration of 1 Gm. a day for six weeks did not destroy vestibular function although there was occasional partial impairment. In cases of generalized hematogenous tuberculosis and tuberculous meningitis, where the fatality rate is very high, treatment should be carried out for ninety days in daily doses of 40 mg. per kg. of body weight, and the disadvantages of drug toxicity and bacterial resistance should be ignored. In meningitis, 50 mg. of the drug should be given intrathecally at twenty-four to seventy-two hour intervals during the ninety days. The value of concomitant administration of the sulfones and para-aminosalicylic acid requires further evaluation. In every patient in whom the use of streptomycin is contemplated, careful consideration should be given to possibility of more critical future episodes. In milder cases it may be wise to administer the drug only for short periods or to withhold it entirely for the time when it may be needed most urgently.—*Streptomycin in the Treatment of Tuberculosis, J. B. Amberson & W. H. Stearns, Ann. Int. Med., August, 1948, 29: 221.—(H. R. Nayer)*

Superinfections during Antibiotic Therapy.—Two patients with pneumococcic pneumonia were given penicillin therapy during the course of which new infections due to gram-negative bacilli appeared. Both patients responded to streptomycin therapy. The mechanism by which the new infection occurs is not clear but it may be due to a disturbance in the degree of bacterial antagonism which is normally present. The importance of frequent bacteriologic examinations before and during treatment with an antibiotic is stressed. It is not advisable to start treatment with a combination of penicillin and streptomycin.—*Occurrence of Superinfections during Antibiotic Therapy, E. Appelbaum & W. A. Leff, J. A. M. A., September 11, 1948, 138: 119.—(H. Abeles)*

Streptomycin in Pregnancy.—Two patients, both in the second trimester of pregnancy, were treated with streptomycin. One patient had tuberculosis of the urinary tract and the other had far advanced pulmonary tuberculosis. Both patients gave birth to normal infants without neural defects. The placenta was normal in both cases.—*Streptomycin Therapy, Effects on Fetus, E. H. Watson &*

R. M. Stow, J. A. M. A., August 28, 1948, 37: 1599.—(H. Abeles)

Chemotherapy for Pulmonary Infections.—The number of postoperative pulmonary infections following major abdominal and thoracic procedures was greatly reduced by inhalation of relatively small doses of micronized penicillin or streptomycin mixtures during the operative and postoperative period. —*Prevention of Postoperative Pulmonary Infections, Inhalation of Micropowdered Penicillin and Streptomycin, G. V. Taplin, S. H. Cohen & E. B. Mahoney, J. A. M. A., September 4, 1948, 138: 4.—(H. Abeles)*

Sulphetrone for Tuberculosis.—Sulphetrone was administered to patients with pulmonary tuberculosis considered unsuitable for other forms of active therapy. The dose was 3 Gm. daily, given in tablets of 0.5 Gm. every four hours for about a week. Thereafter, the daily dose was gradually increased to 5 to 8 Gm. daily. Fluids were limited to 1,500 cc. daily and alkali was given with each dose. The patients were continued on the drug indefinitely. No beneficial effect was noted in cases of tuberculous meningitis, miliary disease or bronchopneumonia. Only one patient with very far advanced disease was benefited. Patients with somewhat less advanced disease often showed considerable improvement. The most frequent toxic effect was the appearance of anemia. Other side effects were headache, nausea, vomiting and rashes.—*Chemotherapy of Pulmonary Tuberculosis with Sulphetrone, T. Anderson & S. J. Strachan, Lancet, July 24, 1948, 2: 185.—(A. G. Cohen)*

Sulphetrone for Tuberculosis.—A total of 44 cases of tuberculosis, of which 42 were pulmonary, were treated with sulphetrone. There was improvement in 22, no change in 5, progression in 6 and death in 11 cases. The improvement was considerable in 9, moderate in 7 and slight in 6. It was not dramatic in any case.—*Chemotherapy of Tuberculosis with Sulphetrone, M. G. Clay & A. C. Clay, Lancet, July 31, 1948, 2: 180.—(A. G. Cohen)*

Sulphetrone for Tuberculosis.—Seventy cases of pulmonary tuberculosis were treated with sulphetrone. Prior to treatment, ferrous sulphate and brewer's yeast were administered for two weeks. The patients were kept in bed. The objective was attainment of a blood level of from 7.5 to 10 mg. per cent. A level greater than 12.5 mg. per cent was dangerous. Because early doses are not well tolerated, these were small and were increased gradually. The drug was given intramuscularly in miliary tuberculosis and tuberculous meningitis. The patients were watched for the appearance of anemia. Minor toxic symptoms were headache, nausea, mental depression and cyanosis. Danger signals were continuous headache, nausea, vomiting, vertigo and mental confusion. Treatment lasted from a few days to eighteen months. There were no beneficial effects in miliary tuberculosis or tuberculous meningitis. Improvement was seen in 12 of 17 cases of acute fibrocaseous tuberculosis and in 13 of 22 chronic cases. Each of 4 cases of primary pulmonary tuberculosis and 6 of 8 cases of strictly exudative disease were improved. In the chronic hematogenous type all 4 cases were improved, as were 3 of 4 cases of productive tuberculosis. In every case, the trend in exudative cases was halted and reversed.—*Treatment of Tuberculosis with Sulphetrone, D. G. Madigan, Lancet, July 31, 1948, 2: 174. —(A. G. Cohen)*

Experimental Chemotherapy in Mice.—A review of the literature on the use of the mouse in the study of experimental tuberculosis convinced the authors that mouse tuberculosis has not been sufficiently delineated for practical purposes. When appropriate allowances are made for several variables, however, a common pattern begins to emerge. It was concluded that: (*1*) the white mouse is more resistant to tuberculous infection than most mammalian species; (*2*) it can be easily and regularly infected with the mammalian and avian varieties of *M. tuberculosis* by a variety of routes; (*3*) the lesions produced are quite diversified and are not histologically

identical with those described for other species; and (*4*) the particular histoarchitecture observed depends upon the variety, virulence and dose of infecting organism, the route of inoculation, and the time of observation. It is apparent that the histopathologic picture produced by a given set of the foregoing variables must be standardized before its alteration by any therapeutic agent can be evaluated.—*The Use of Mice in Experimental Chemotherapy of Tuberculosis: I. Rationale and Review of the Literature. G. W. Raleigh & G. P. Youmans, J. Infect. Dis., May–June, 1948, 82: 197.*—(*F. G. Petrik*)

Experimental Tuberculosis in Mice.—The evolution of mouse tuberculosis under standardized experimental conditions is described. The clinical and histopathologic pattern is defined and it is felt that potential therapeutic agents can be recognized by the alterations they induce in this pattern. *The Use of Mice in Experimental Chemotherapy of Tuberculosis: II. Pathology and Pathogenesis. G. W. Raleigh & G. P. Youmans, J. Infect. Dis., May–June, 1948, 82: 205.*—(*F. G. Petrik*)

Experimental Chemotherapy in Mice.—Twenty potential chemotherapeutic agents were administered to tuberculous mice. Under aseptic precautions 0.1 mg. of virulent human type tubercle bacilli was injected intravenously into mice of the Strong A strain weighing between 20 and 25 Gm. The compounds were administered to the mice by admixture with the diet, except streptomycin which was given subcutaneously. The compounds tested were selected either on the basis of *in vitro* bacteriostatic tests or on the basis of known chemotherapeutic action. Administration of each drug was begun two to three days before the animals were infected with tubercle bacilli and was continued with all compounds, except streptomycin, for twenty-eight days. Streptomycin treatment was started on the day of infection and extended over 35 days. Treated mice were compared in every respect with homologous control animals infected at the same time. Of the 20 compounds tested only streptomycin and para-aminosalicylic acid were found to be appreciably effective. It is felt that survival time, weight change and the histopathologic picture constitute a triad of useful measures in the assay of any chemotherapeutic agent and that of these three determinations the latter is most important.—*The Use of Mice in Experimental Chemotherapy of Tuberculosis: III. The Histopathologic Assay of Chemotherapeutic Action, G. P. Youmans & G. W. Raleigh, J. Infect. Dis., May–June, 1948, 82: 221.*—(*F. G. Petrik*)

Aerosol Antibiotic Administration.—The efficacy of nebulization therapy depends on the local effect of the antibiotic in the bronchial tree and is not dependent on the absorbtion of the antibiotic by the blood. Penicillin "aerosol" therapy is indicated for infections of the tracheobronchial tree caused by bacteria which are sensitive to penicillin; streptomycin "aerosol" is indicated for those infections caused by streptomycin-sensitive organisms. Penicillin and streptomycin "aerosols" have proved to be of greatest value in the eradication of purulent exudates complicating bronchiectasis. The method has been of particular value in the preoperative preparation of patients for lobectomy. "Aerosols" of penicillin or streptomycin are sometimes helpful in the treatment of nontuberculous lung abscess. Their value is limited by the difficulty in getting the drug into the diseased portion of the lung. Nebulization therapy may be helpful in treating patients with asthma, emphysema, and chronic bronchitis only if these conditions are associated with infections of the bronchial tree caused by bacteria sensitive to the antibiotic used. "Aerosols" are of no value in treatment of respiratory infections caused by viruses.—*Administration of Antibiotic Preparations by the Aerosol Method: A Critical Evaluation, A. M. Olsen, Medical Clinics of North America, July, 1948, 32: 1077.*—(*L. Hyde*)

Postural Changes in Vital Capacity.—The vital capacities were determined in 9 adult

male subjects in the standing and supinepositions. Cuffs placed at the bases of the arms and legs were inflated to diastolic pressure, above systolic pressure, or left uninflated. If the cuffs were inflated to a level above the systolic blood pressure with the subject standing, the reflux of venous blood from the trunk and head did not lower vital capacity when he reclined. There was no difference between the vital capacity of a subject standing with the cuffs inflated above the systolic pressure and the vital capacity of the same subject standing with noninflated cuffs.—*Postural Changes in Vital Capacity with Differential Cuff Pressures at the Bases of the Extremities, G. S. Campbell & R. B. Harvey, Am. J. Physiol., March 1, 1948, 152: 671.*—(*G. C. Leiner*)

Residual Air.—The residual lung volume was determined in 6 trained subjects with the "nitrogen dilution method" in which the nitrogen in the residual air is diluted by re-breathing a known volume of oxygen, and by two new methods. (*1*) On the "helium substitution method" the residual air of the lungs is replaced by a known helium-oxygen mixture. The helium is then washed out of the lungs by inhalation of air or oxygen and its volume measured. (*2*) In the "volume expansion method" measurements are made of the increase in gas volume which results from the expansion of the residual air when the ambient pressure is rapidly reduced from 4 to one atmospheres. The mean residual air in the 6 subjects as determined by the nitrogen dilution method was 1,491 cc., by the helium substitution method 1,427 cc., by the volume expansion method 1,429 cc.—*Residual Lung Volume Determinations by the Methods of Helium Substitution and Volume Expansion, T. L. Willmon & A. R. Behnke, Am. J. Physiol., April 1, 1948, 153: 188.*—(*G. C. Leiner*)

Intra-alveolar Pressure.—The roentgenographic appearance of the lungs is dependent to a great extent upon the degree of filling of the pulmonary vessels. With an increase in filling, the lung pattern becomes more prominent; with a decrease, it becomes less prominent. The intra-alveolar pressure, by influencing the filling of the vessels, affects the appearance of the lungs. Variations in the intra-alveolar pressure also produce an effect upon the circulation and, in consequence, upon the heart. During routine roentgenography of the lung, there may be marked variations in the intra-alveolar pressure. Laurell has shown that roentgenograms are comparable only when the same intra-alveolar pressure is present each time the lung is examined. The author used a manometer attached to a mouthpiece to measure intra-alveolar pressure and made roentgenograms at different pressures. The volume of the heart increases one-third when the alveolar pressure is low and decreases one-third when it is high. In normal persons a positive pressure of 40 cm. of water is needed to compress visibly the central branches of the pulmonary artery and this pressure corresponds to the intravascular pressure in the pulmonary artery. Thus a rough estimate of the pressure in the pulmonary arteries can be obtained by determining the intra-alveolar pressure at which the arteries in the hilum are reduced in size. Though this may be a rough method of determining the pulmonary arterial pressure, it is of practical importance for the determination of the degree and the prognosis of mitral valvular disease and may also be of value in the diagnosis of other diseases causing increased blood pressure in the pulmonary artery. A high intra-alveolar pressure, compressing the central branches of the pulmonary artery, aids in demonstrating the hilar lymph nodes on the chest roentgenogram. In a study of primary tuberculosis, hilar node enlargement was demonstrable in 62 per cent of the cases. In a later series of cases studied with both normal and high intra-alveolar pressures, an enlargement of the hilar nodes was found in 97 per cent of the cases. In pulmonary stasis the widened vascular shadows and the opacities caused by pulmonary edema decrease considerably with high intra-alveolar pressure; in some cases the appearance of

stasis may be entirely obliterated while in others it becomes less pronounced. In cases of acute bronchopneumonia the diffuse, opaque network of the parenchyma appears thinner and the opacity is less pronounced but more sharply defined in films made at high pressure. Altering the intra-alveolar pressure causes little or no change in the roentgenographic appearance of acute lobar pneumonia, acute and chronic atelectasis, chronic pneumonia and in most cases of tuberculosis. Cases illustrative of the various types of pulmonary disease discussed are presented.—*The Importance of Intra-alveolar Pressure in the Diagnosis of Pulmonary Diseases, N. Westermark, Radiology, May, 1948, 50: 610.—(G. F. Mitchell)*

Pulmonary Edema.—The intracranial pressure was increased in dogs up to over 300 mm. of mercury. This did not cause pulmonary edema. However, a condition resembling pulmonary edema may be produced in such experiments due to the aspiration of saliva.—*An Attempt to Produce Pulmonary Edema by Increased Intracranial Pressure, A. Surtshin, L. N. Katz & S. Rodbard, Am. J. Physiol., March 1, 1948, 152: 589.—(G. C. Leiner)*

Pulmonary Arterial Blood Pressure.—In anesthetized cats, measurements of the pressure in the pulmonary artery were made by means of a cannula inserted into the pulmonary artery. In some instances there were indications of a nervous influence on the pulmonary arterial pressure. Inhalation of carbon dioxide produced increase in the pressure; oxygen want had a similar but stronger effect. Oxygen inhalation produced a drop of the pressure. Increase of inspiratory or expiratory resistance increased the pulmonary arterial pressure. The blood flow through any part of the lungs depends on its ventilation, the flow shifting from badly ventilated parts of the lungs to parts which are well ventilated. —*Regulation of Pulmonary Arterial Blood Pressure, G. Liljestrand, Arch. Int. Med., February, 1948, 81: 162.—(G. C. Leiner)*

Pulmonary Vascular Diseases.—The causes of peripheral vascular disease of the lung may be grouped as follows: (*1*) intrinsic disease of the vessel wall or obstruction of the lumen; (*2*) changes in vessels produced by adjacent disease of the pulmonary parenchyma; and (*3*) toxic and hypersensitivity states resulting in increased permeability of vessel walls. Arteriosclerosis, arteriolosclerosis and pulmonary hypertension cause changes in the pulmonary vessels. Obliterative vascular disease regularly accompanies advanced pulmonary emphysema. Obstruction of the vascular lumen may be due to a great variety of conditions: foreign material, septic, organic, or inorganic fragments circulating in the systemic blood vessels, neoplasms, blood dyscrasia, and parasitic infestations. Vascular changes are produced by the adjacent disease in acute pneumonia, in chronic inflammatory conditions and neoplasms, in tuberculosis and in silicosis. Some pathologic lesions are associated with a great variety of conditions: poisoning with sulfonamide compounds, serum sickness, eczema, lupus erythemathosus, acute rheumatic fever, and periarteritis nodosa. If sensitization becomes profound, organic destruction of the vessel wall occurs with necrosis. The effects of exogenous and endogenus toxins on the walls of small vessels are identical with the manifestations of hypersensitivity. Vascular permeability may be altered in long standing protein deficiency, in beri-beri, and in a peculiar iron deficiency described by Waldenstrom.—*Peripheral Vascular Disease in the Lung, Roentgenologic Manifestations, R. P. Barden & D. A. Cooper, J. A. M. A., June 12, 1948, 137: 584.—(H. Abeles)*

Laryngeal Swab in Pulmonary Tuberculosis.—In 201 patients with radiological evidence of tuberculosis but with no sputum or with no demonstrable tubercle bacilli in the sputum, cultures were made of gastric contents and laryngeal swabs. Gastric lavage was slightly superior for obtaining cultures of tubercle bacilli from inpatients; there was no difference in the results of the 2 methods for

outpatients. Laryngeal swabs are preferred by patients and are less laborious for physicians, nurses and laboratory technicians.—*Laryngeal Swab in Early and Convalescent Cases of Pulmonary Tuberculosis, G. B. Forbes, B. J. D. Smith, J. V. Hurford & V. H. Springett, Lancet, July 24, 1948, 2: 141.*—(*A. G. Cohen*)

Tuberculin Testing.—Tuberculin testing will be very important in the proposed BCG inoculation programs, since it will be necessary to determine which persons are to be vaccinated and to follow those who have been vaccinated. The only immediate and tangible evidence of any change in the immune mechanism is the reversal of the tuberculin reaction from negative to positive. There is a tendency for the cutaneous sensitivity to tuberculin to disappear in cases of healed tuberculosis. The frequency of this reversal is definitely related to the degree of sensitivity exhibited by the patient on the original test. Of 2,490 persons with positive tuberculin tests who were followed for five to fifteen years, 11 per cent lost their tuberculin sensitivity. This occurred much more frequently in persons whose sensitivity was low initially, and most often in children; a similar reversal was observed rarely in adults. Clinical evidence has been accumulated to show that a tuberculous patient who has overcome his infection may completely lose his cutaneous sensitivity but retain his acquired resistance to the disease. Immunity against tuberculosis in such a patient is different from the immunity of a person who has never been infected and who reacts negatively to intradermal tuberculin. Animal experiments support this thesis. Statistical conclusions drawn from tuberculin surveys made twenty or thirty years ago are no longer valid. New country-wide surveys of varied adult groups are indicated. The tuberculin skin test is becoming an important aid in general diagnosis especially when one deals with puzzling diseases of the thorax.—*Changing Concepts in Tuberculin Testing, G. G. Stilwell, Medical Clinics of North America, July, 1948, 32: 1095.*—(*B. Hyde*)

Tuberculosis in Students.—Since 1929 the University of Minnesota Students' Health Service has tested all medical students with tuberculin and made chest roentgenograms of all reactors. In 1936 over 65 per cent of the nonreactors on entrance became reactors while in school. Following the introduction of a control program, this percentage fell to 3.2 per cent in 1947. There was no significant difference in the development of the chronic reinfection type of tuberculosis among those entering as reactors and those who became reactors while in school.—*Prevention of Tuberculosis Among Students of Medicine, H. S. Diehl, R. E. Boynton, S. Geist-Black & J. A. Myers, J.A.M.A., September 4, 1948, 138: S.*—(*H. Abeles*)

THORACOPLASTY IN THE TREATMENT OF PULMONARY TUBERCULOSIS[1,2]

An Analysis of Results Five to Twenty-six Years after Operation

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INTRODUCTION

Extrapleural thoracoplasty has occupied a role of increasing importance over the years in the surgical treatment of pulmonary tuberculosis. The value of the procedure in situations where it is definitely indicated has never been seriously challenged and the early results obtained have been eminently satisfactory. Because of the treacherous nature of tuberculosis and the tendency for its victims to undergo relapses and exacerbations of the disease from time to time, it was felt that a careful analysis of the late results obtained in a large series of patients, carefully controlled and accurately followed for many years, might reveal much information of value in guiding the future handling of this disease.

OBSERVATIONS

Glen Lake Sanatorium, a public institution of 640 adult bed capacity, caring for the tuberculous individuals of Hennepin County, including Minneapolis, was first opened in 1916 and has been in continuous operation for the past thirty-two years. During the period of this study, 1916 through 1942, a total of 6,394 patients suffering from pulmonary tuberculosis were admitted to this institution for treatment. This group included 3,773 patients with far advanced disease, 1,978 with moderately advanced disease, and 693 with minimal tuberculosis. The senior medical and surgical staff has, with one or two exceptions, remained almost unchanged for the entire period. Thus the factors of indications, selections of patients, preoperative and postoperative care and the like, which might influence the series, have remained remarkably constant throughout except as altered by accrued experience.

The first thoracoplasty operation at Glen Lake Sanatorium was performed on January 22, 1922 by the late Dr. Arthur A. Law who pioneered through the early portion of the series. He remained the surgical director until early in 1928 and on his retirement was succeeded by one of the writers (T. J. K.). Since the fall of 1940 another two of the writers (V. K. F. and L. M. L.) have carried on the surgical work. During the study, a total of 613 patients, or 9.6 per cent of the patients with pulmonary tuberculosis admitted to the institution from 1916 to 1943, were subjected to thoracoplasty. Six hundred and nine of these patients had unilateral thoracoplasty carried out in 1,562 operations, while four had

[1] From the Glen Lake Sanatorium, Oak Terrace, Minnesota.

[2] Presented before the Medical Section, as part of the symposium on *Surgery—Trends—Late Results*, at the 44th Annual Meeting of the National Tuberculosis Association, New York, New York, June 16, 1948.

bilateral thoracoplasties performed in 12 operations, making a total of 1,574 (including 88 revisions) operations for the entire series, an average of 2.6 operations per patient (tables 1, 2, 3).

Sex, Age and Side of Operation

The series included 270 males and 343 females, a distribution which was comparable with that found in the general population of the Sanatorium. The age range for the males was from eleven to sixty-four years and for females from sixteen to sixty-six years. Eighteen of the patients were between ten and twenty years of age, 213 between twenty and thirty, 206 between thirty and forty, 121 between forty and fifty, 42 from fifty to sixty, while 13 were beyond the age of sixty at the time surgery was undertaken. In 295 of the patients the thoracoplasty was performed on the right side, in 314 the operation was on the left, and in 4 the procedure was bilateral. Twelve and one-tenth per cent of the patients

TABLE 1

Stage of disease at time of thoracoplasty, 1922 to 1948

STAGE OF DISEASE	SEX OF PATIENTS			ADMISSIONS ADULT-PULMONARY 1916 TO 1943
	Male	Female	Total	
Far advanced	176	211	387	3,773
Moderately advanced	93	132	225	1,978
Minimal	1	0	1	643
Total	270	343	613	6,394

Thoracoplasty 613 = 9.6 per cent of admissions.

operated upon presented unilateral disease at the time of their surgery, while 87.9 per cent had bilateral involvement, though usually the contralateral process was not recently active or progressive. At the time of the thoracoplasty operation contralateral pneumothorax was present in 27 patients, contralateral paraffin pneumonolysis in 3, and contralateral thoracoplasty in 4.

Selection of Patients

Prospective candidates for thoracoplasty were always presented before a staff conference by the physician upon whose service the patient was in residence. At this time a complete review of the history, physical examination, laboratory findings, and all significant roentgenograms was presented, together with the physician's evaluation of the situation and recommendations as to treatment or requests for aid. In all situations the evaluation of the attending physician in closest contact with the patient was duly honored and his recommendations were given careful consideration. In general, patients with acute or recent disease were not deemed suitable for thoracoplasty, the emphasis being placed on the selection of the "good chronic" for surgical treatment, though there were many deviations from this general plan. No patient who had a reasonable chance of obtaining a satisfactory result from surgical treatment was denied

surgery because of being considered to be a "bad risk" or because of the presence of tuberculous or nontuberculous extrapulmonary complications.

TABLE 2

Total thoracoplastics, 1922 to 1943 (excluding 88 reoperations)

Number of operations per patient	Unilateral thoracoplasty					
	Known Deaths		Others		Total	
	Patients	Operations	Patients	Operations	Patients	Operations
1 Operation	45	45	54	54	99	99
2 Operations	79	158	157	314	236	472
3 Operations	51	153	153	459	204	612
4 Operations	24	96	37	148	61	244
5 Operations	1	5	6	30	7	35
6 Operations	2	12			2	12
Total	202	469	407	1,005	609	1,474
	Bilateral thoracoplasty					
1 Operation each side	1	2			1	2
1 Operation 1 side, 2 on other			2	6	2	6
1 Operation 1 side, 3 on other			1	4	1	4
Total	1	2	3	10	4	12
Grand total	203	471	410	1,015	613	1,486

TABLE 3

Type of operation and side

	With anterolateral		Without anterolateral		Total
	Right	Left	Right	Left	
Complete	42	53	48	70	213
Partial	76	69	129	122	396
Total	118	122	177	192	609

Exclusive of 4 with bilateral

Indications

By far the greatest number of patients subjected to thoracoplasty over the years have been operated upon because of the presence of pulmonary cavitation,

either large or small, and because of the persistence of phenomena arising from the cavity, such as continued cough with discharge of tubercle bacilli, or pulmonary hemorrhage. A smaller group of patients were operated on because of primary pyothorax or pyopneumothorax complicating previous attempts to control the disease by artificial pneumothorax. A few patients were subjected to extrapleural thoracoplasty for other indications, *e.g.*, the obliteration of an extrapleural empyema pocket, to intensify the collapse of a previously unsuccessful extrapleural pneumolysis, to relieve traction, displacement or discomfort produced by the atelectatic retracting of an upper lobe tuberculosis, or to relieve bronchial kinking. In the majority of the patients operated upon attempts at the induction of artificial pneumothorax had either failed or the treatment had proved unsatisfactory. A few primary thoracoplasties without previous attempt at pneumothorax were carried out even many years ago under special circumstances.

Three hundred and eighty-three of the patients (172 males and 211 females) were operated upon because of far advanced disease, 229 (97 males and 132 females) because of moderately advanced disease, and one male had a complete thoracoplasty because of a minimal tuberculous lesion with pulmonary hemorrhage. The stage of disease mentioned indicates the stage of disease at the time the surgery was undertaken, not at the time of admission to the sanatorium and has been made according to the present National Tuberculosis Association classification of disease as evidenced on the preoperative roentgenograms. These films were completely re-examined by the writers in the preparation of this paper. Such a reclassification has been necessitated by the changes of classification which have been made on two or three occasions since the inception of this series.

Type of Operation

The earlier portion of this series was represented by a group of patients in whom the Wilms-Sauerbruch type of limited posterior paravertebral resection was carried out. Almost invariably the lower stage procedure was done first if multiple stage operations were carried out. There was but one complete single stage operation in the series and the patient died within twenty-four hours. It was considered mandatory at this time to perform a phrenic nerve interruption preliminary to thoracoplasty. As the inadequacy of the limited resections in patients with extensive disease gradually became evident, wider and wider segments of rib were removed until the operation approached the Brauer subscapular type of thoracoplasty. By the spring of 1927 a change was made so that an upper posterior thoracoplasty was usually done first and the complete posterior thoracoplasty divided into three stages instead of two. Anterolateral thoracoplasty was used as early as 1926 and adopted more frequently after Welles' original article on anterolateral thoracoplasty was published in 1927 (2). At first it was used almost invariably as a sequence to complete posterior thoracoplasty. Later it was combined with a partial thoracoplasty and soon came to be used as the second stage of the series with the upper posterior first and the intermediate posterior third. The use of the lateral or anterolateral operation as a first stage was not deemed advisable and, therefore, not used in this series.

The interval between operations in the earlier portion of the series was excessively long, at times even a matter of several months. Gradually, however, as the series developed, an interval of approximately three weeks between stages was adopted and was utilized in the majority of the cases. More recently this interval has been reduced to two weeks.

All of the earlier thoracoplasties were carried out in an extremely rapid time "with one eye on the clock" in an attempt to complete the operation before shock supervened. Records as low as nine minutes from incision to last stitch of closure for a lower stage six rib thoracoplasty, and eleven minutes from incision to last stitch of closure for an upper stage five rib thoracoplasty were made. Needless to say, excessive blood loss, long residual vertebral rib stumps, perforation of the

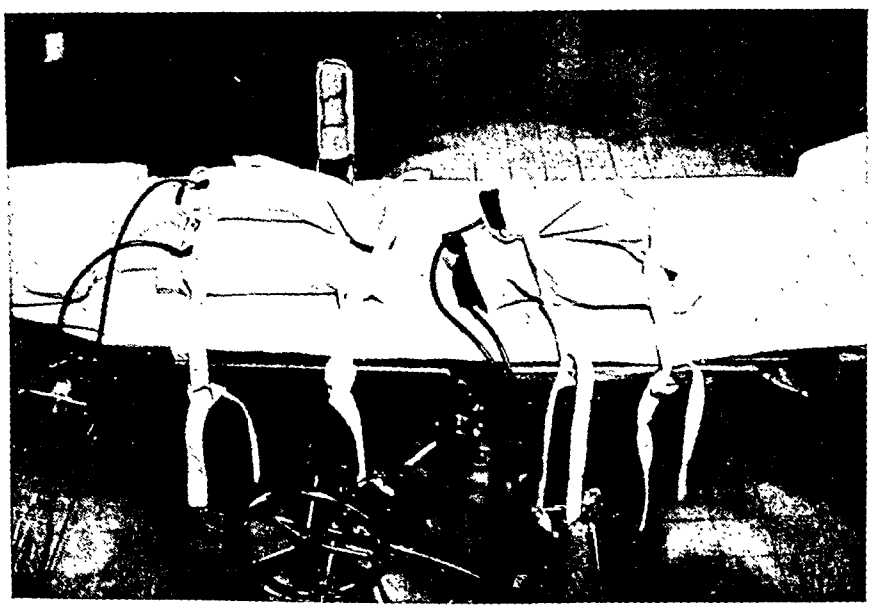

FIG. 1. Balloons used in posturing patient on the operating table.

pleura, and severe shock were frequently encountered. With improvements in technique and particularly with a change to local infiltration as the anesthesia of choice in 1929, the emphasis on speed was discarded and a more careful, meticulous type of thoracoplasty carried out, resulting in improved postoperative condition of the patients. While some of the earlier operations were carried out in the direct lateral position, a change was soon made to a three-quarters prone position which has been used consistently ever since. This position may be seen in figures 1 and 2, which illustrate the balloons used in posturing the patient and the patient in operative position upon the table.

Blood loss in the earlier thoracoplasty operations was not calculated, but is known to have been excessive. Calculations of the average blood loss for nearly 100 thoracoplasty stages carried out under local anesthesia were reported in 1937 (1) as follows: upper posterior thoracoplasty three to four ribs, 450 cc. blood loss; intermediate posterior thoracoplasty three to four ribs, 296 cc.; lower

posterior thoracoplasty three to four ribs, 196 cc.; and anterolateral thoracoplasty three to four ribs and cartilages, 250 cc. These values were obtained by calculation of the hemoglobin content of all of the sponges and packs used during the course of the operation and all of the dressings used by the patients in the first forty-eight hours after surgery and the result was then expressed in terms of cubic centimeters of blood of a hemoglobin content at the patient's preoperative hemoglobin level.

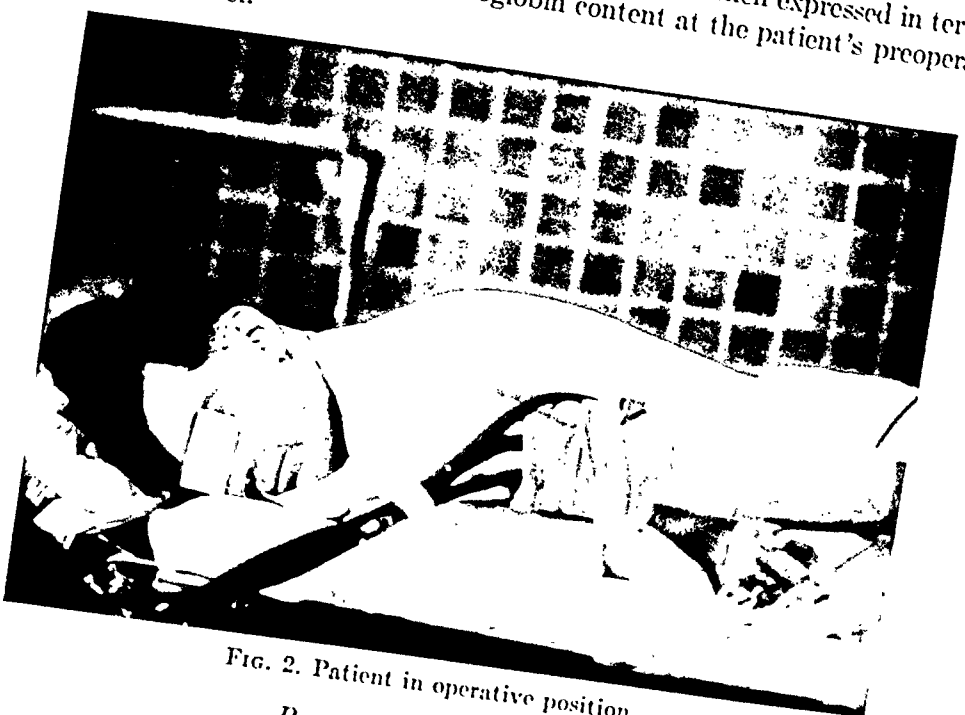

FIG. 2. Patient in operative position.

Preoperative Preparation

The preoperative preparation of the patient was usually as simple as possible in an effort to disturb the patient's normal routine to a minimal degree. A careful survey of all factors was always made so that the risk might be evaluated and all necessary precautions taken to protect the patient in every possible way. Subsequent to 1926, the cardiovascular status of all prospective thoracoplasty patients was carefully studied by the attending cardiologist, Dr. Olga S. Hansen, and electrocardiographic studies were made before operation, between stages, and thereafter. A special effort was always made to ensure that the patient had raised his usual quota of sputum before surgery was undertaken. For this reason for many years the majority of the thoracoplasties were carried out in the afternoon when the patient has usually completed his daily cavity drainage.

If the operation was to be carried out under local anesthesia, the patient was given 0.1 Gm. of nembutal at two hours and one hour before surgery, followed by a hypodermic of 0.010 Gm. of morphine or pantapon at approximately one half hour before going to the operating room. An additional hypodermic of 0.060 Gm. of codeine was sent to the operating room with the patient to be used at any time when unusual apprehension, nervousness or other symptoms might appear during the course of the surgery.

The original operations in the series were done entirely under nitrous oxide anesthesia, but very soon paravertebral block with novocain followed by nitrous oxide anesthesia became the routine anesthetic. Through 1928 and 1929 occasional cases were done under local anesthesia alone. With increasing experience, local anesthesia became the anesthetic of choice and thereafter was used almost exclusively for operations which could be carried on under this type of anesthesia alone. In no instance was it necessary to change to a general anesthesia when this sequence had not been anticipated before surgery was undertaken. Nitrous oxide, ethylene, or cyclopropane were administered in addition to the local anesthesia in a few instances. The general impression that under local infiltra-

FIG. 3. Support of chest while patient is encouraged to cough.

tion anesthesia the patients came through their surgery in better condition with less shock, less blood loss and in better general condition has been adequately confirmed over the years. The ability of the patient to cough and expectorate during, as well as immediately following, surgery has been a distinct advantage, particularly in the patient with larger amounts of sputum.

Postoperative Care

Postoperatively there are a number of points which were considered important to the safety and comfort of the patient. In order to avoid aspiration of material into the good lung, care was taken never to turn the patient or allow the patient to turn so that the uninvolved lung was dependent while awake or asleep. Opiates were used in dosage sufficient to control distress but not to destroy the cough reflex. The patient was required to cough and expectorate sputum at intervals from the time of surgery, using expectorants and proper support to the deribbed chest wall, as illustrated in figure 3. Fluids were routinely administered

postoperatively by retention enema and by the intravenous or subcutaneous routes when necessary. Transfusions were used on but few occasions in the whole series. Patients were permitted and encouraged to take fluids and food by mouth as soon as tolerated. Many patients were able to take a reasonably normal meal within a few hours after the operation.

If any paradoxical chest wall motion was noted, sandbags were immediately applied to the area or if necessary the chest was firmly strapped with adhesive tape over rubber sponges. Local area dressings were changed as necessary. Penrose drain tubes which were used routinely were removed in approximately forty-eight hours. Patients were encouraged and urged to move around as soon as possible after surgery. They were routinely required to sit up for dressings the morning following the operation and subsequently at frequent intervals. Early motion of the arm and shoulder on the operated side was insisted upon and every attempt was made to keep the patient from feeling that the operation would in any way cripple him. Because surgery alone does not cure tuberculosis, all patients were maintained on strict bed-rest for a period of at least six months following the completion of all stages of the thoracoplasty procedure.

Mortality

Twenty-two of the 613 patients in the entire series died within fourteen days of the operation (table 4). This represents a mortality rate of 3.59 per cent per patient and 1.40 per cent per operation. A total of 34 patients died within eight weeks of the time of operation, yielding an eight weeks' mortality of 5.55 per cent per patient or 2.16 per cent per operation (Table 5). One patient died upon the operating table and two others died during the first twenty-four hours following surgery, apparently from shock. Three died on the second day following operation, one on the third, 2 on the fourth, 3 on the fifth, 2 on the seventh, one on the eighth, 2 on the ninth, 2 on the tenth, and 2 on the eleventh day after surgery. Eleven of the deaths may justly be attributed to an extension of the tuberculosis. Two of the patients definitely aspirated material through a bronchial fistula to cause the spread of the disease. An additional patient, considered at postmortem examination as having died of asphyxia, probably also should have been classified as a tuberculous death. Two, who on the death certificates were listed as pneumonia, were proven at postmortem examination to have nontuberculous pneumonias. One patient was listed as dying of pulmonary edema, one from right heart failure and one on postmortem examination as tuberculous myocarditis and rheumatic endocarditis, making a total of 3 patients apparently dying of cardiac conditions. One died from a cerebral vascular accident, probably resulting from low blood pressure and thrombosis, one from a pulmonary embolism on the eighth postoperative day, and one patient from an accidental pneumothorax produced on the fourth postoperative day by an attempt to refill a contralateral pneumothorax which was present at the time of surgery. The cause of death in 15 of this group was established at necropsy.

Eleven patients died after the first stage operation, 6 after the second, 1 after the third, and one after the fourth operation. Twelve deaths occurred after

TABLE 4

Thoracoplasty series mortality, 1922 to 1948

DEATH SUBSEQUENT TO OPERATION	
8 weeks	34
8 weeks to 4 years (inclusive)	92
5 to 9 years	55
10 to 14 years	17
15 to 19 years	5
Total	203
SURVIVAL AFTER LAST OPERATION (ANALYSIS MARCH 1948)	
1 to 4 years	8
5 to 9 years	127
10 to 14 years	99
15 to 19 years	60
20 years and over	20
Total	314
SURVIVAL AFTER LAST OPERATION (RECENT INFORMATION UNOBTAINABLE MARCH 1948)	
1 to 4 years	43
5 to 9 years	38
10 to 14 years	12
15 to 19 years	3
Total	96

TABLE 5

Thoracoplasty series mortality, 1922 to 1948

	PER CENT OF PATIENTS	PER CENT OF OPERATIONS
OPERATIVE DEATHS		
2 Weeks	3.60	1.40
2 Months	5.55	2.16
CONDITION AT TIME OF LAST FOLLOW-UP		
Alive	410	66.88
Dead from all causes	203	33.12
Unrelated Cause	34	5.55
Tuberculosis	169	27.57

local anesthesia, 7 after nitrous oxide and local, 2 after ethylene and one following spinal anesthesia. There were 11 males and 11 females. The age groups were quite representative of the age groups in the whole series, with only 2 of the deaths occurring in patients over 45 years of age, and 2 under the age of 20.

An additional 12 patients died of the following causes within six weeks or two months of operation: 8 from pulmonary tuberculosis; one from tuberculous meningitis; one from pulmonary embolism; one from chest wall flutter; and one from cardiovascular failure with peripheral edema. One of the tuberculous deaths in this group also resulted from aspiration of pus from a pyopneumothorax cavity through a bronchial fistula. The cause of death in 7 of this group was confirmed by postmortem studies.

A graphic representation of how the operative mortality has decreased with the development of the series may be seen in figure 4. Each vertical column in the figure represents the number of operations (and the number of patients sub-

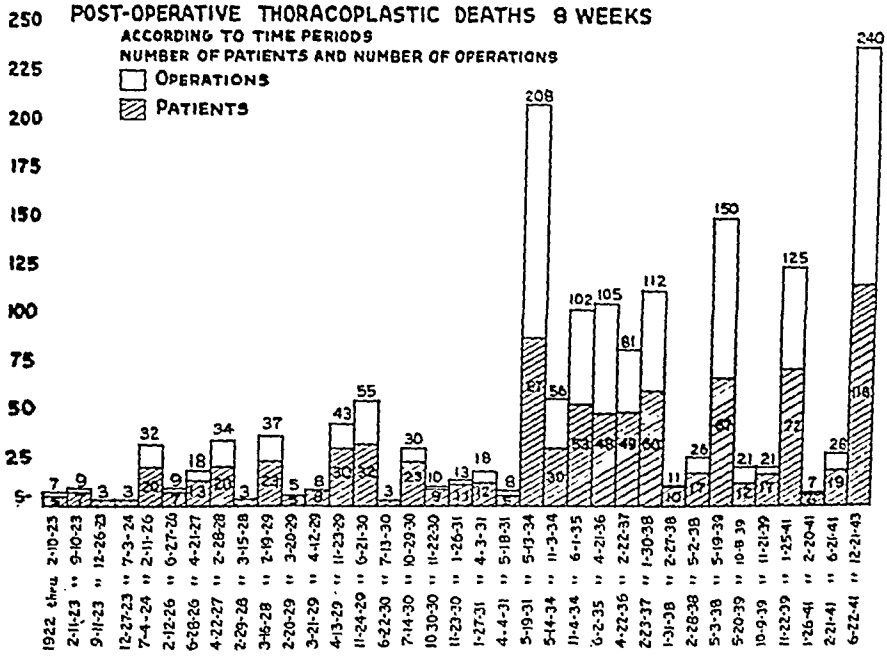

FIG. 4. Postoperative thoracoplastic deaths 8 weeks.

jected to surgery) which were performed during the various periods of time which elapsed between any two postoperative deaths. All deaths, regardless of cause, which occurred during the eight week period following operation are included. The death which interrupted the series on May 13, 1934, after 208 consecutive operations on 87 patients, was caused by an accidental pneumothorax on the contralateral side induced in an attempt to refill a pneumothorax pocket present before operation. The operative mortality (within eight weeks) since 1931 has been 3.0 per cent per patient and 1.16 per cent per operation. It is interesting to note that 59 per cent of the mortality within eight weeks has occurred in the first 21 per cent of the series.

Shock was a very prominent feature in all of the earlier operations and was frequently of severe degree. It is very difficult from the study of charts alone to obtain any accurate indications of the frequency of this condition because of the

great variation in its degree and in the interpretation of what constitutes shock. Three patients definitely died in this state. In the latter portion of the series shock became a factor of much less importance and in a severe form was rarely, if ever, seen.

Blood transfusions were rarely used in this series which was carried out before the days of blood banks with blood freely available. It is quite probable that in the early portion of the series especially the use of blood transfusions would have been a distinct advantage. They have not been necessary in the latter portion of the series, but might definitely have hastened the recovery of some patients and possibly offered a chance for a shorter interval between stages than the one customarily used.

Major wound infections were not of frequent occurrence and records of only 35 or 2.2 per cent can be found listed. Perforation of the pleura was noted not infrequently in the early portion of the series where an extremely rapid surgical resection was carried out. In the latter portion of the series, however, with a more careful operative technique, perforations have rarely occurred. Tuberculous sinuses resulting from damage to lung, pleura or to tuberculous lymph nodes have been recorded in only 6 patients in the whole series.

Scoliosis of any degree whatsoever as noted on the roentgenogram has been reported in 165 patients or 26.9 per cent of the entire series, but in only 13 (2.12 per cent) was it either moderate or marked. It was seen most frequently in young people, particularly in young girls of poor musculature. Scoliosis can be produced, however, by destruction of portions of the erector spinae group of muscles or their nerve supply, or by resection of transverse processes which represent the only remaining site of muscle attachment after removal of the ribs. No planned program for prevention and treatment of scoliosis was used in this series as it was deemed unnecessary because of the low incidence and slight degree of this complication.

Spread of Disease

A spread or exacerbation of tuberculosis was encountered 94 times in 90 patients in this series which represents a rate of 14.66 per cent per patient or 6.34 per cent per operation. Thirty-four "spreads" occurred in areas not previously involved with tuberculosis, while 60 occurred in areas in which there had previously been tuberculous disease. The latter group raises the question of how much of the total figure represents exacerbation of previously existing disease. This question cannot be definitely answered and it is probable that most of the new lesions represent a bronchiogenic spread of disease. In only 10 of the patients was the "spread" on the same side as the thoracoplasty. It is noteworthy that one-half of the patients who developed an extension of disease on the same side had a paralyzed diaphragm on that side at the time of surgery.

Thirty-eight of the patients who developed extension of disease were operated upon for large cavities, 29 for small cavities, 17 for pyopneumothorax, and 6 for miscellaneous causes. It is interesting to note that 5 of the patients with pyopneumothorax who developed a spread of disease had a definite bronchial fistula

present at the time of surgery. The hazard of a bronchial fistula in the patient with a pyopneumothorax cannot be too strongly emphasized. Sixty-two of these patients were operated upon under local anesthesia, 21 under gas and local infiltration, 3 under ethylene, one under nitrous oxide alone, one under spinal anesthesia, and one under ethylene and local anesthesia, and one under an anesthetic of unstated type. The incidence of "spread" after local anesthesia was 4.9 per cent, while under general anesthesia it was 8.4 per cent. Many other factors besides anesthetic alone must be considered in the development of a bronchiogenic spread of tuberculosis following thoracoplasty. Important among these are medications before and after operation, the ability to expectorate, the cooperation of the patient, his position in moving, and his position in bed. Consequently any correlation of "spread" to anesthesia is not an exact one. A spread of disease occurred after the first operation in 50 instances, after the second in 24, after the third in 16, after the fourth in 3 and once after the fifth operation. Among the 90 patients who developed a spread of tuberculosis following surgery, 11 died within the first two weeks after surgery, and 7 within the subsequent six weeks. Thus a total of 18 or 20 per cent died within two months. Eleven more patients died within the first year, 10 within the second year, and 19 at times varying from three to thirteen years after surgery. The spread of tuberculosis postoperatively was definitely a causative factor in the subsequent death of the majority of these individuals who died within two years.

RESULTS

Four hundred and ten, or 66.8 per cent of the entire series operated upon, still survive for a period of five to twenty-six years following surgery, giving a series mortality for all causes of 33.12 per cent. If the 34 patients who died at various times up to eighteen years following surgery from causes totally unrelated to tuberculosis are deducted from the total number of patients now dead, it may be seen that 27.57 per cent of the entire group of 613 thoracoplasty patients eventually died, either early or late, from tuberculosis or its complications. Among the causes of death unrelated to tuberculosis are: carcinoma, 9 patients; suicide, 6; accidents, 5; nontuberculous appendicitis and peritonitis, 3; cardiovascular complications, 3; psychosis, 2; nontuberculous pneumonia, 2; mesenteric thrombosis, one; meningococcic meningitis, one; nephritis following scarlet fever, one; and operation for a nontuberculous condition, one. As near as can be determined, the tuberculosis was under control at the time of death in all of these patients. These survival figures and the accompanying tables clearly indicate the stability of these lesions in these individuals once the tuberculosis has been controlled by means of extrapleural thoracoplasty.

Cavity Closure and Reversal of Infectiousness

In 88.1 per cent of the patients whose sputum contained tubercle bacilli prior to surgery, the organisms subsequently disappeared from the sputum, according to the direct examination technique used at the sanatorium over the years covered by the study. Undoubtedly, cultures of sputum or cultures of gastric washings

or guinea pig inoculations would alter this percentage materially but, as these procedures were not in use consistently over the years, they were not considered in this study where a uniform listing was desirable. As may be seen in table 6, of the patients who presented large cavities (more than 4 cm. in any one diameter prior to surgery), closure was obtained in only 49.6 per cent, with disappearance of tubercle bacilli from the sputum in 76.5 per cent. In the patients with small cavities (table 7), the incidence of cavity closure was considerably greater, namely

TABLE 6

Cavity closure and "sputum conversion"
(Large cavities)

	TOTAL	SPUTUM			
		Positive Before		Negative Before	
		Negative after	Positive after	Negative after	Positive after
Closed	70	58	8	4	0
Open	71	43	23	5	0
Total	141	101	31	9	0

49.6 per cent cavity closure.
76.5 per cent "sputum conversion."

TABLE 7

Cavity closure and "sputum conversion"
(Small cavities)

	TOTAL	SPUTUM			
		Positive Before		Negative Before	
		Negative after	Positive after	Negative after	Positive after
Closed	218	136	4	77	1
Open	99	55	15	28	1
Total	317	191	19	105	2

68.7 per cent cavity closure.
91.0 per cent "sputum conversion."

68.7 per cent, with "sputum conversion" in 90.95 per cent. Further analysis of the late results of the patients operated upon because of large and small cavity, with particular reference to mortality, both early and late, in relation to cavity closure and "sputum conversion" is not complete and will be published at a later date.

Pyopneumothorax

Ninety-four patients were operated upon because of pyopneumothorax. In this group, obliteration of the empyema pocket was obtained in 61 and question-

able closure in one additional patient, or 67.3 per cent of the patients operated upon for this condition. Failure of closure occurred in 32.7 per cent of those who survived the surgery. In this entire group, 5 died within two weeks with empyema pocket unclosed, one under eight weeks with the empyema pocket unclosed, and 6 additional patients within one year, 2 with the empyema pockets obliterated and 4 unobliterated. Twenty-five additional patients of this series died after periods longer than one year following the surgery, and of these 25, obliteration of the empyema pocket had failed to occur in 11, while 13 such cavities were definitely closed and one questionably closed at the time of death. Fifty-nine and seven-tenths per cent of the entire group still survive. The presence of a bronchial fistula in this group proved a distinct hazard for 5 of the group of 17 developed a contralateral spread of disease following surgery.

Supplementary operations to the standard thoracoplasty were frequently necessary in this series of empyemas, a complication which as a rule demands a much more extensive thoracoplasty and more numerous stages than uncomplicated pulmonary tuberculosis. Catheter drainage, rib resection for open drainage, unroofing operations, and plastic closures were all utilized in various patients in this group in an attempt to bring about obliteration of the pleural cavity. As many as seven or eight separate operations were required in some patients. Increased experience in handling empyemas has produced better results. Catheter drainage at the base laterally has been considered mandatory in all patients with definite or suspected bronchial fistulae or secondary infection. Moreover, with chemotherapeutic agents available, catheter drainage has been used with increasing frequency and to advantage in patients with straight tuberculous pyonpeumothorax without secondary infection. Closed drainage with or without irrigation or continuous suction has been of distinct value in removing all fluid from the pocket and reducing its size and bringing about more rapid obliteration of the pleural space during the course of the collapse procedures. A more complete report of this group will be made in a future communication.

Revised Operations

A series of 63 patients have been subjected to various types of supplementary and revision operations with and without packs in attempts to improve the results when the primary surgical procedures had not been entirely satisfactory. In general, these procedures were well tolerated and aided materially in improving the clinical result. Cavity closure occurred in 56.6 per cent and "sputum conversion" occurred in 73 per cent of this group of 63 patients.

SUMMARY

1. Thoracoplasty was performed on 613 tuberculous patients (1,474 individual operations) during the twenty year period 1922 to 1943.

2. Sixty-six and eight-tenths per cent of the patients are alive five to twenty-six years after operation; 27.57 per cent ultimately died of tuberculosis or its complications.

3. The operative mortalities were 3.59 per cent per patient (1.4 per cent per

operation) during the two weeks after surgery. During the eight weeks after operation the mortality was 5.55 per cent per patient and 1.16 per cent per operation.

4. Since 1931 the postoperative mortality (eight weeks) has been 3 per cent per patient and 1.16 per cent per operation.

SUMARIO

La Toracoplastia en el Tratamiento de la Tuberculos Pulmonar: Análisis de los Resultados de Cinco a Veintiséis Años después de la Operación

1. En el veintenio 1922–1943 se ejecutó la toracoplastia en 613 tuberculosos (1,574 distintas operaciones).

2. De estos enfermos, 66.8 por ciento se hallan vivos de cinco a veintiséis años después de la operación; 27.57 por ciento murieron por fin de tuberculosis o de las complicaciones de la misma.

3. La mortalidad operatoria representó 3.59 por cien enfermos (1.4 por cien operaciones) en las dos semanas consecutivas a la intervención. Durante las ocho semanas consecutivas a la intervención la mortalidad se elevó a 5.55 por cien enfermos y 1.16 por cien o operaciones.

4. Desde 1931 la mortalidad postoperatoria (ocho semanas) ha sido de 3.0 por ciento en enfermos y de 1.16 por ciento en operaciones.

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SIGNIFICANT ELECTROCARDIOGRAPHIC CHANGES FOLLOWING CHEST SURGERY[1, 2]

Electrocardiographic Changes Currently Considered Indicative of Serious Heart Disease Occurring Postoperatively in Patients with Complete Absence of Clinical Manifestations of Heart Disease

SOLOMON M. RAUCHWERGER AND FREDERICK A. ERSKINE

INTRODUCTION

The limits of normal electrocardiographic patterns have not been completely defined as yet. During World War II many studies were made at induction centers and military installations by competent investigators and numerous new variations of the normal pattern were reported (1). On the basis of this fact alone, it has been emphasized repeatedly that a diagnosis of myocardial damage should not be made solely on the basis of electrocardiographic findings.

A correlation study of the electrocardiographic findings and subsequent clinical course of 112 patients who were subjected to chest surgery has been undertaken. The surgical procedures were performed during a five year period from 1943 to 1948 at a 1,000-bed tuberculosis sanatorium and electrocardiograms were obtained before and after operation in all of the patients studied. It was noted early in the study that T-wave changes in Lead I and Lead IV, usually interpreted as signifying the presence of severe myocardial damage (3, 8, 9), occurred with notable frequency without subsequent clinical evidence of heart disease.

Because of this significant feature and also because there is relatively little information in the literature concerning electrocardiographic changes following thoracoplasty and other forms of chest surgery, the findings are reported. The benign course followed by these patients and the amount of additional surgery well tolerated after the T-wave changes occurred suggest that a revision of the electrocardiographic criteria for inoperability may be in order. Three patients included in this series did develop the electrocardiographic and clinical findings characteristic of well-known, specific disease states and these cases are discussed in detail below.

METHODS AND MATERIALS

The patients comprising this series, with the exception of 4 with bronchiectasis and 2 with pulmonary abscess, all had pulmonary tuberculosis for which surgery had been performed. There were 93 thoracoplasties, 42 on the left side and 51 on the right; 9 pneumonectomies, 3 primary and 6 secondary to a thoracoplasty; and 12 lobectomies, 8 primary and 4 secondary to a thoracoplasty. The excision cases combined with a thoracoplasty were considered as thoracoplasties in tabu-

[1] From the Department of Medicine and Surgery, Veterans Administration Oteen, North Carolina.

[2] Published with permission of the Chief Medical Officer, Department of Medicine and Surgery, Veterans Administration, who assumes no responsibility for the opinions expressed or conclusions drawn by the authors.

TABLE 1

Table of T-wave changes following various forms of chest surgery

TYPE OF COLLAPSE	NUMBER OF PATIENTS	T_1 (I)	T_1 (L)	T_1 (D)	T_2 (I)	T_2 (L)	T_2 (D)	T_3 (I)	T_3 (L)	T_3 (D)	T_4 (I)	T_4 (L)	T_4 (D)	TOTAL NUMBER OF CHANGES
Thoracoplasty right	51	0	8	2	0	5	0	5	3	4	2	2	1	32
Thoracoplasty left..	42	7	12	6	2	5	1	4	2	2	6	2	5	54
Lobectomy.....	8	0	2	0	0	0	0	1	0	2	1	1	1	8
Pneumonectomy....	3	1	1	2	0	0	0	1	0	0	1	2	1	9
Lucite pack.....	8	0	3	1	0	1	0	4	1	1	0	0	1	12

(I) = inversion.
(L) = low or isoelectric.
(D) = diphasicity.

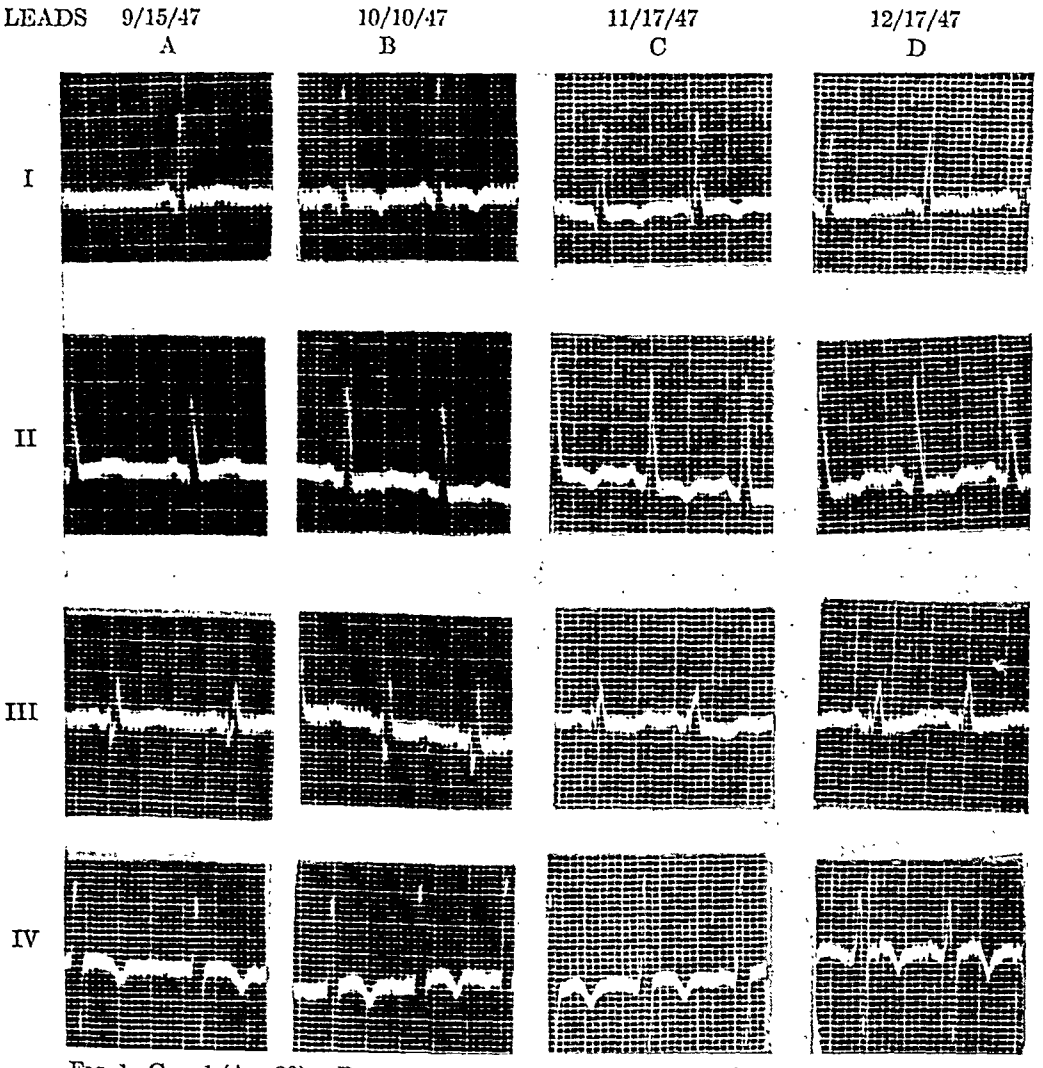

FIG. 1. Case 1 (Age 26). Roentgenogram revealed bilateral pulmonary tuberculosis with cavitation on the left. Previous collapse therapy consisted of a left pneumothorax, left phreniclasia, and pneumoperitoneum. A three-stage left thoracoplasty was performed between September 22 and December 3, 1947. Cyclopropane and ether were the anesthetic agents employed. Blood pressure was labile ranging from 110/80 to 140/100. The electrocardiographic pattern observed occurred following the first-stage thoracoplasty.

(A) Before thoracoplasty—T_4 negative; (B) after first stage—negative T_1; diphasic T_2; negative T_4; (C) before third stage—negative T_1; diphasic T_2, T_3; negative T_4. (D) After completion of thoracoplasty—negative, low T_1; negative T_4.

lating results as no significant data resulted from further breakdown. There were 8 cases of extrapleural pneumonolysis with plombage (methyl methacrylate), 3 alone, and 5 combined in some form with a thoracoplasty.

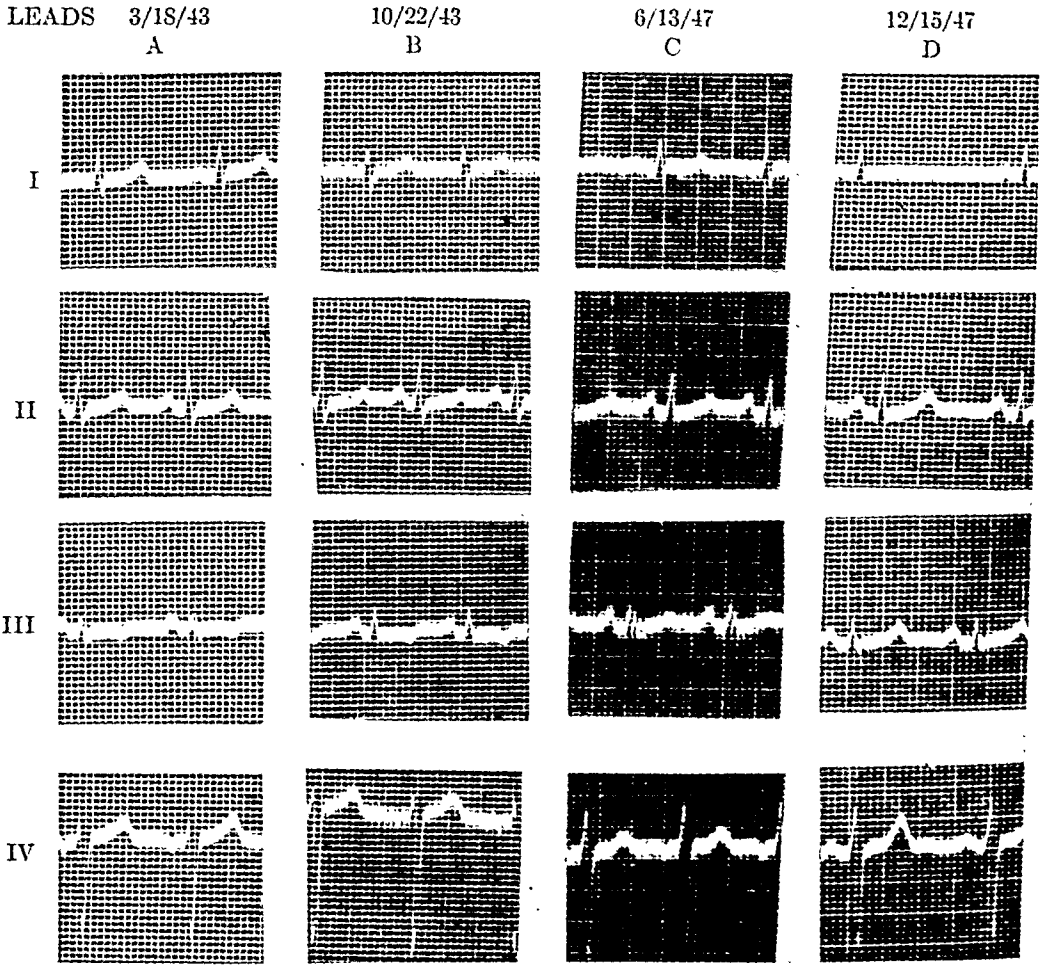

FIG. 2. Case 2 (Age 55). Chest roentgenogram revealed extensive fibro-exudative disease with cavitation on the left and a fibrotic lesion on the right. This patient had a three-stage left thoracoplasty including a revision thoracoplasty performed between November 5, 1943, and September 22, 1944. On June 17, 1947, a left pneumonectomy was done. Again cyclopropane and ether were the anesthetic agents. The electrocardiographic changes seen in tracing dated December 15, 1947 were first noted June 20, 1947 but the tracing for this date could not be located.

(A) Before surgery—normal tracing; (D) Following pneumonectomy—T_1 isoelectric.

Each patient had a complete cardiac examination, including an electrocardiogram, before and after operations. The electrocardiographic study included the four standard leads (I, II, III, and CF IV) in all cases, and in the doubtful cases the six classical chest leads were also employed. In addition, an electrocardiogram was made prior to each stage of thoracoplasty. The examinations also

included a clinical evaluation of the patient on the morning surgery was to be performed.

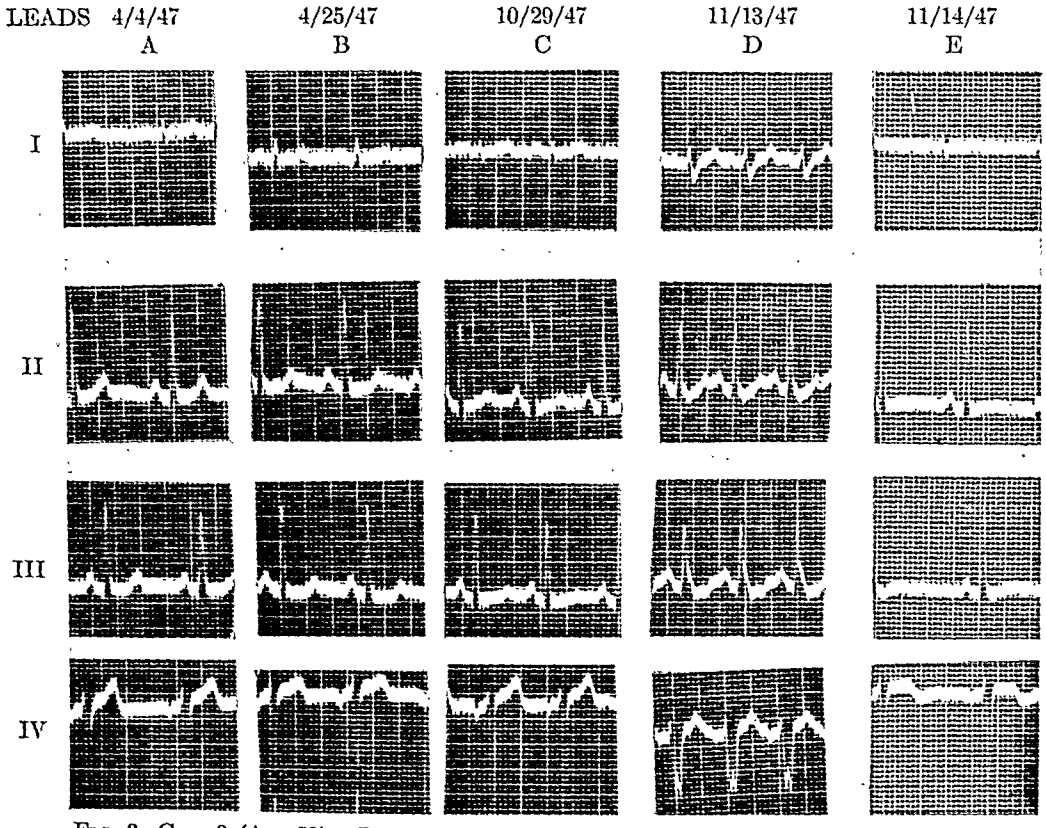

FIG. 3. Case 3 (Age 52). Patient was admitted with a diagnosis of far advanced pulmonary tuberculosis with cavitation on the right. A three-stage right thoracoplasty was done between April 7 and May 13, 1947. On November 12, 1947, he had an excision of an osteomyelitic rib. Approximately twenty-four hours later, he developed signs and symptoms of a pulmonary embolism on the left side, manifested by sudden onset of severe pain in the chest, more marked on the left, and accompanied by evidence of shock. Anticoagulants and supportive measures were administered and recovery was uneventful. An electrocardiogram on November 13, 1947, revealed the findings characteristic of pulmonary embolism. The electrocardiographic changes gradually resolved over the ensuing several days.

(A) Before surgery—normal pattern; (B), (C) following thoracoplasty—no change; (D) after excision of osteomylitic rib—deep S_1; depressed ST_2, ST_3; (E) 24 hours after (D)—low, slurred QRS_1 and poor T-waves.

RESULTS

Pronounced T-wave changes in Leads I and IV were noted in 34 of the 112 cases reviewed. In 18 there was marked lowering (to less than 100 microvolts) or inversion of T_1 alone. In 16 cases there occurred a combination of lowered or inverted T_1 with a similar change in T_4. The T-wave changes in Leads II and III were as tabulated in table 1.

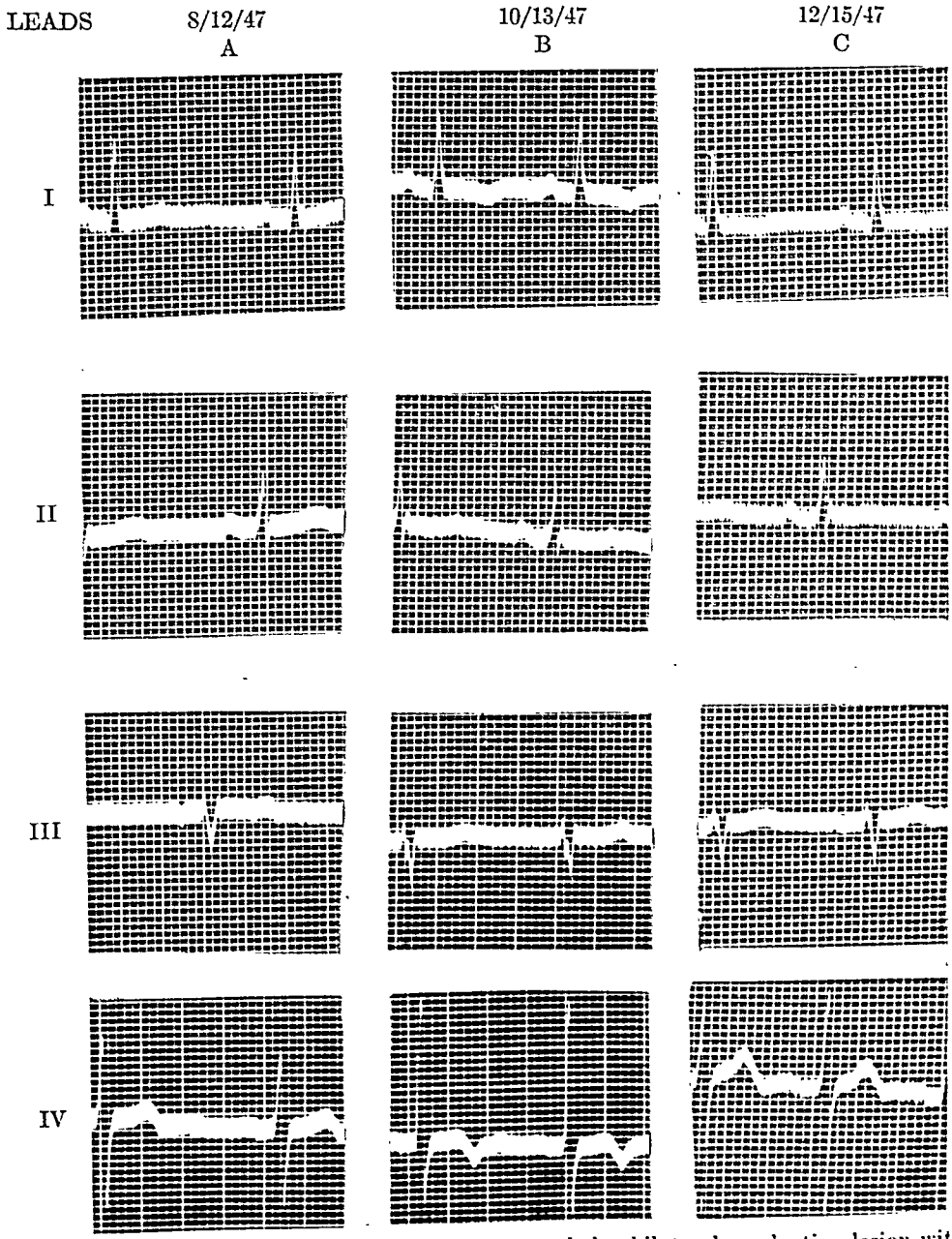

FIG. 4. Case 4 (Age 24). Roentgenogram revealed a bilateral productive lesion with some degree of atelectasis of the left lung. A two-stage left thoracoplasty was performed between September 15 and October 20, 1947. Cyclopropane and ether were the anesthetic agents used. Changes in the electrocardiographic pattern occurred after the first-stage thoracoplasty.

(A) Before surgery—left axis deviation; (B) following first stage—inverted T_1 and T_4; (C) after completion of thoracoplasty—low, inverted T_1; T_4 now upright.

Other changes occurring less frequently (not statistically significant) were: (a) tendency for increase in amplitude of P-waves *without* evidence of right heart strain in the vast majority of instances; (b) elevation in S-T segment in 4 cases, in Leads II, III and IV; (c) frequent extrasystoles (especially prominent following 2 pneumonectomies); and (d) occasional splitting of QRS_4. In review of the records of 23 patients who had a phrenemphraxis with or without pneumoperitoneum or pneumothorax, the only unusual findings were inversion of T_4 in two (table 1), and inversion of T_1 in one.

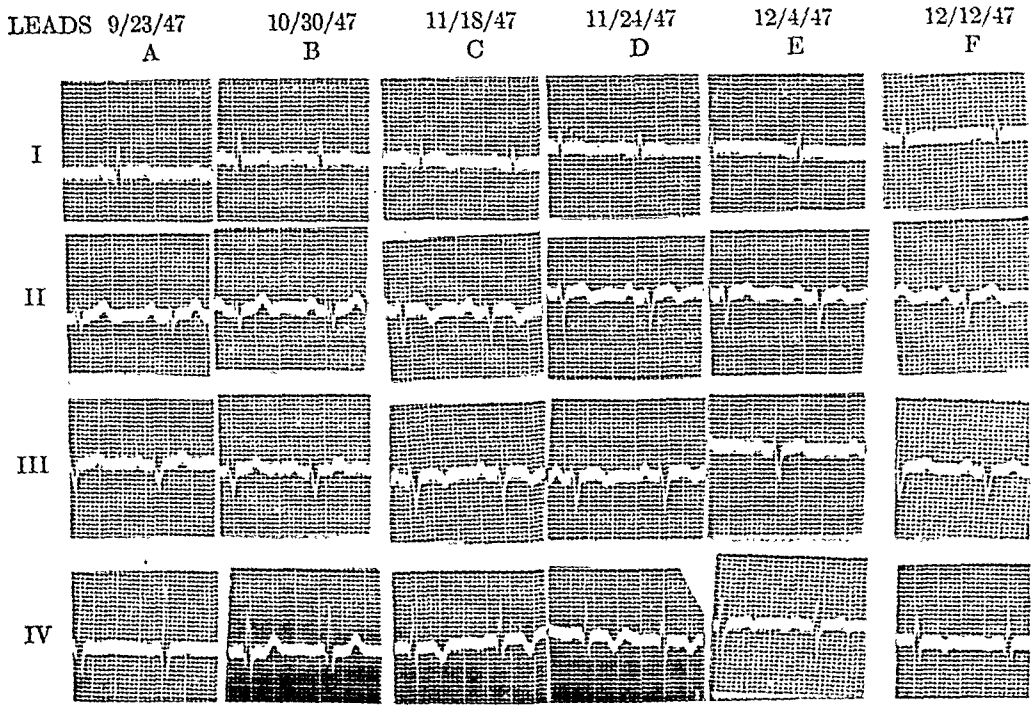

FIG. 5. Case 5 (Age 46). Roentgenogram revealed bilateral pulmonary tuberculosis of a fibro-caseous nature with cavitation on the right. Previous collapse therapy consisted of an unsuccessful right pneumothorax, a right phreniclasia, and pneumoperitoneum. On October 17, 1947, an anterior first-stage right thoracoplasty, and on October 31, 1947, a posterior right thoracoplasty were performed. On November16, 1947, the patient experienced an acute episode of pain in the left side of the chest which gradually increased in intensity, especially on deep inspiration. An electrocardiographic tracing taken on Novebmer 18, 1947, revealed a pattern compatible with that of an acute pericarditis. Frequent examinations failed to reveal evidence of a friction rub or a pericardial effusion. Subsequent tracings showed resolution of the acute process and on December 16, 1947, a second-stage posterior thoracoplasty was performed under cyclopropane and ether. Between January 16 and February 2, 1948, he had a two-stage cavernostomy with no untoward effect. Serial electrocardiograms up through February 4, 1948, revealed normal findings.

(A) Before surgery—left axis deviation; (B) before second stage thoracoplasty—no change; (C) T-waves inverted in all leads; (D) six days after (C)—T waves diphasic; (E) T_2, T_3 now upright; T_1 and T_4 low and diphasic; (F) T_1 isoelectric; T_4 inverted.

The presence of a negative T-wave in Lead I, especially in conjunction with a

negative or diphasic T-wave in Lead IV (figure 1), if persistent, is currently considered strongly suggestive of myocardial damage (3, 8, 9). Alterations in Leads II, III, and CF IV alone may be produced by shifting of the mediastinal contents without coexistent heart damage (figure 3). This is equally true

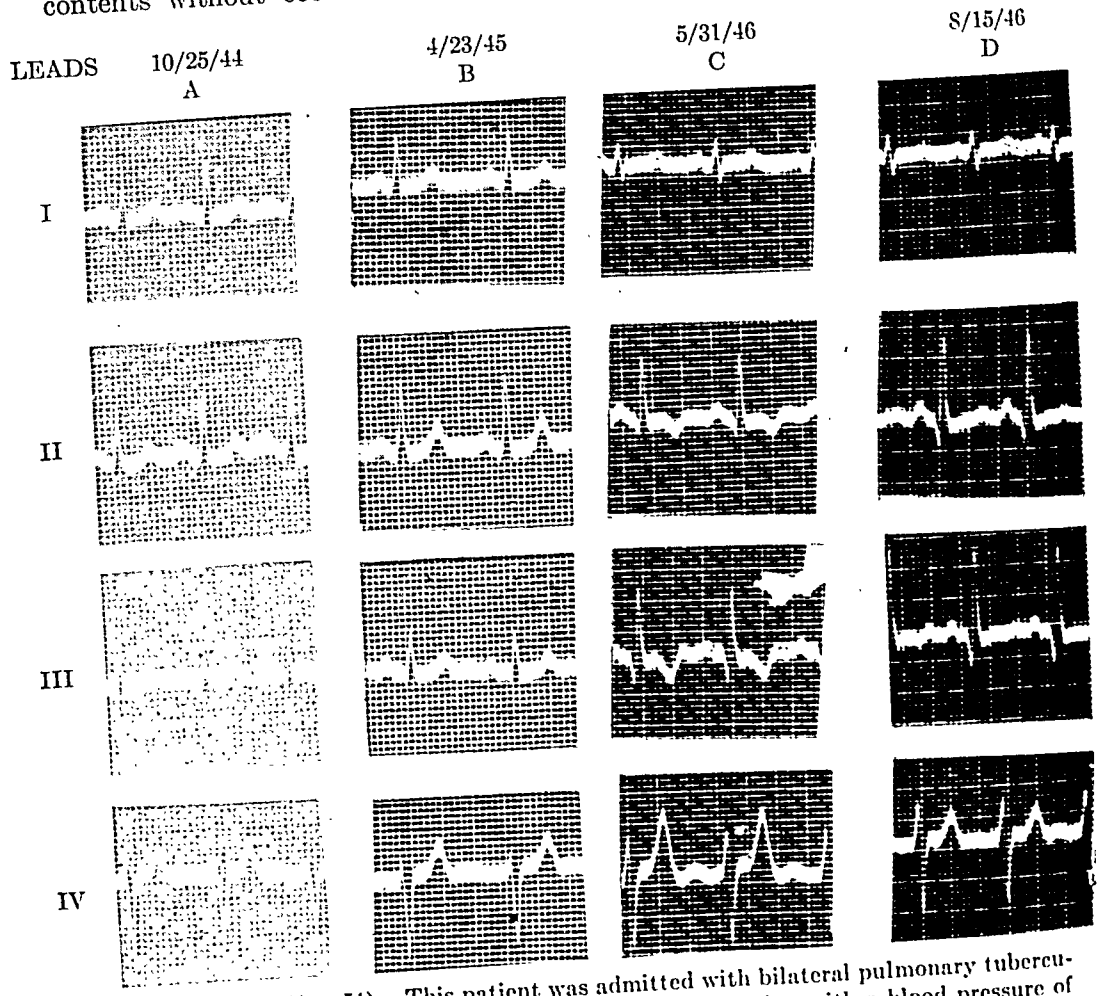

FIG. 6. Case 6 (Age 54). This patient was admitted with bilateral pulmonary tuberculosis with cavitation on the left, and a borderline hypertension with a blood pressure of 150/100. A three-stage left thoracoplasty was done between October 1944, and April 1945. On May 30, 1946, he developed substernal pain of a severe nature with radiation to the left arm. An electrocardiogram taken at this time revealed a tracing compatible with that of a posterior myocardial infarction. (A) Before surgery—normal tracing; (B) following surgery—no change; (C) Q_2T_2, Q_3T_3 pattern (posterior occlusion); (D) Q_2 and Q_3 persistent; T_2 upright; T_3 diphasic.

whether the cause be effusion, adhesions, position of the patient, or surgical measures, and is not considered indicative of heart disease *per se*. This fact has been reported and verified many times (1, 4, 5, 6, 11).

In the 10 cases in the series in which there was shifting of the mediastinum, there were only 2 in which the T-waves in Lead I became of low amplitude, and in

only one was the T-wave in Lead I inverted. Changes in posture from recumbency to sitting did not alter the observed patterns in Leads II, III, and IV in these patients (13). As noted in table 1, however, over half of the group (62.5 per cent) did reveal changes postoperatively in Leads II, III, or IV, either alone or in combination. The T-wave changes observed in Leads I and IV, however,

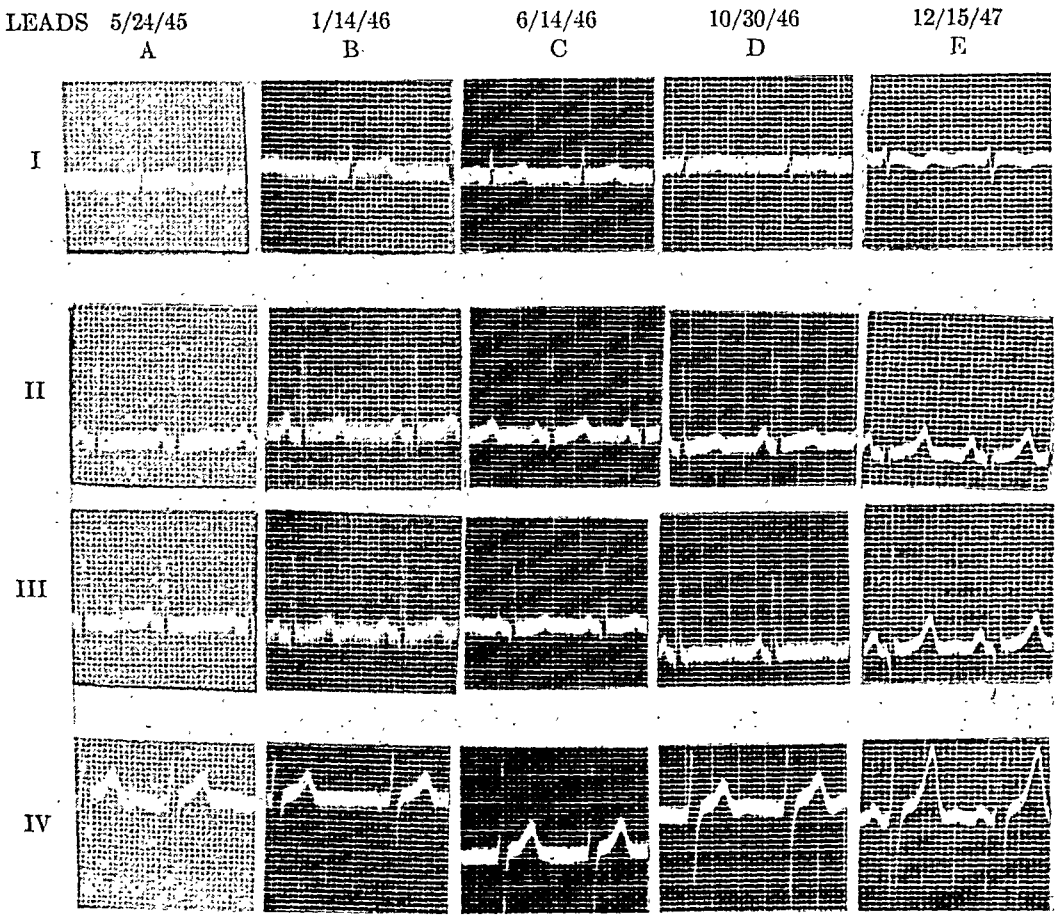

FIG. 7. Case 7 (Age 26). Roentgenogram revealed fibro-exudative disease with cavitation on the left. A two-stage left thoracoplasty was performed between April 13 and May 4, 1945. Electrocardiographic changes were first noted on October 30, 1946. Subsequently this patient had a revision thoracoplasty on November 1, 1946. He had a left lower lobectomy January 1, 1946.

(A), (B), (C), (D) Following 2-stage thoracoplasty—normal tracing; (E) following revision of thoracoplasty—T_1 inverted; T_4 shows high amplitude.

were of paramount concern as they were so striking, tended to persist, and yet did not appear to be due to myocardial damage.

During the course of the study, whenever T_1 inversion was found to have occurred, either alone or in combination with an altered T_4, a careful scrutiny was made for any clinical evidence of heart disease. This included a search for cor-

onary insufficiency, any marked variation in blood pressure, change in heart sounds or rhythm, diminution in cardiac reserve, and especially for evidence of cor pulmonale. These procedures were carried out in an attempt to account for the electrocardiographic changes in Leads I and IV on the basis of demonstrable organic heart disease. It is felt that if heart disease of a nature serious enough to produce the electrocardiographic changes noted were actually present, some clinical evidence thereof would have manifested itself during the early postoperative stages, or certainly within the follow-up period.

Although changes in many instances were pronounced when first noted, they were not considered sufficient to contraindicate subsequent chest surgery. The early experience in this question was obtained by observation of the satisfactory postoperative course followed by several patients for whom surgery was absolutely necessary despite electrocardiographic changes. As already mentioned, these later stages of surgery were well tolerated from a cardiovascular standpoint and there appears to be no justification for assuming the presence of heart disease merely on the strength of electrocardiographic changes which have not been described in the absence of heart disease prior to this time (2, 7).

TABLE 2

T-wave changes subsequent to phrenemphraxis in twenty-three patients

T-WAVE CHANGE TYPE OF COLLAPSE	T_1 (I)	T_1 (L)	T_1 (D)	T_2 (I)	T_2 (L)	T_2 (D)	T_3 (I)	T_3 (L)	T_3 (D)	T_4 (I)	T_4 (L)	T_4 (D)	TOTAL NUMBER OF CHANGES
Phrenemphraxis right	1	0	2	1	0	0	0	2	0	1	0	1	8
Phrenemphraxis left	0	0	2	0	0	0	0	0	1	1	0	1	5

(I) = inversion.
(L) = low or isoelectric.
(D) = diphasicity.

DISCUSSION

The reasons for these T-wave changes are not known at present, just as the reasons for inversions of T_2 and T_3 were not known when this phenomenon was first observed in normal individuals. There is no adequate justification for assuming that any toxins accompanying the tuberculous process should suddenly cause severe heart damage with the electrocardiographic changes as noted. Nor were there any postoperative pneumonias or spreads of disease, such as have been reported as possible etiologic factors by Thomson *et al.* (12), which occurred to account for the T-wave changes. The drugs used postoperatively included morphine sulfate; barbiturates, as hypnotics; multiple vitamins; penicillin and streptomycin. It is known that none of these drugs with the possible exception of streptomycin, will produce the changes recorded. Although investigation of the toxic side effects of streptomycin is far from exhausted, a review of the electrocardiograms of over 200 patients who have received the drug has revealed no

similar changes correlated with its administration. Furthermore, only approximately 50 per cent of patients in whom the electrocardiographic changes were found received streptomycin.

The conditions in the present study are far from analogous to those which existed during the early formative years of electrocardiography. Also the type of patient studied is very dissimilar from the "normals" utilized in so many of the earlier studies. The group in the present study consisted predominantly of seriously ill patients who were subjected to major surgery. Such a select group might well be expected to develop some form of heart strain or permanent myocardial damage. Yet this did not occur or, at least, was not evident in a clinically recognizable degree. Thus it seems reasonable to postulate primarily mechanical "extracardiac" etiologic factors as responsible for the electrocardiographic changes observed. Among these may be included obstructive circulatory phenomena, change in position of the heart with and without compression, and postoperative adhesions. In the two cases in the present series which came to autopsy and were available for study, no adhesions were found between the pleura and the pericardium or between the pericardium and the heart. It is recognized that this small number of cases does not constitute a basis for any definite conclusions in this regard.

It has been suggested by Levinson *et al.* (5, 11) that the presence of air in the chest after pneumothorax might well be the cause for the T-wave changes noted in their series, while Simon and Baum (4) postulate that adhesions comprise the significant etiologic factor in producing ventricular predominance. In the present series the only air in the chest of the patients was occasional postoperative subcutaneous emphysema and pneumothorax was not present. It was observed, however, that in those cases in which there was axis deviation (table 3), a definite shift or rotation of the heart and the mediastinum had taken place.

Although some of the electrocardiographic patterns suggest that of a coronary occlusion, none is typical and the argument that a "silent" coronary attack occurred is considered unlikely, primarily on the following basis:

1. There were no signs of coronary insufficiency, even with subsequent major surgery.
2. In general, patients were in the middle of the third decade or early fourth decade and therefore were far less likely to have a coronary occlusion than an individual in an older age group.
3. Of greatest significance, further check with chest leads failed to corroborate the possibility of coronary occlusion.

If the T-wave changes in Leads I and IV were due to an acute coronary insufficiency precipitated during surgery as a result of massive blood loss, they should have been of a transient nature with reversal to normal as soon as there was an elimination of the precipitating factors (10). Multiple transfusions during and after surgery (which was performed under cyclopropane and ether anesthesia, thus permitting optimum oxygenation) coupled with careful postoperative care including the administration of oxygen, certainly served to correct any cardiac anoxia which may have existed during surgery.

Although no careful neuropsychiatric evaluation was made to elicit any deep-

seated emotional conflicts that might have been present, the average patient was not of an abnormally tensive nature, nor possessed of any profound anxiety state which could conceivably contribute to some of the changes noted. If anything, "spes phthisica" was the prevalent attitude.

Only after evaluation of the manifold considerations discussed above did the writer feel justified in accepting the initial impression *i.e.*, that the electrocardiographic changes in Leads I and IV, ordinarily interpreted as signifying organic heart disease, were occurring and persisting in complete absence of myocardial damage as more than a mere hypothesis. At the very least, in the interest of a conservative, open-minded approach, it may be stated that these electrocardiographic changes occurred in the absence of "clinical and other laboratory evidence" of organic heart disease.

It is hoped that these findings will serve to stimulate further investigation along these lines. Recent work at this institution is continuing to afford support to the above formulation. The opportunity of studying such a large group is

TABLE 3

Axis deviation occurring after chest surgery

(Each case with a deviation of axis showed a concurrent shift of the mediastinum)

TYPE OF COLLAPSE	NUMBER OF CASES	RAD	LAD
Thoracoplasty left	10	2	8
Thoracoplasty right	10	5	5
Lobectomy	8	1	0
Pneumonectomy	3	1	1

RAD = right axis deviation.
LAD = left axis deviation.

definitely unique, and the current policy of including an electrocardiographic tracing as part of the routine admission examination record at this hospital is expected to bear fruit over the next several years. Further correlative studies on this series now in progress include amplitude measurements of various phases of the complexes and tabulation of P-wave changes. These findings will be reported subsequently.

SUMMARY

1. A correlation study was attempted between the clinical course and the electrocardiograms made before and after chest surgical procedures in 112 patients.

2. T-wave changes in Leads I and IV, of a type usually considered to indicate severe heart damage, were found to be present in 30 per cent of the entire series of patients. The changes, consisting of either an inversion of T_1 alone or in conjunction with an inverted T_4, occurred in the complete absence of demonstrable heart disease.

3. Arguments are offered to refute the assumption of the presence of coexistent myocardial damage in these patients.

4. A plea is made for further investigation of the changes recorded.

SUMARIO

Significativas Alteraciones Electrocardiográficas Consecutivas a la Cirugía Torácica

1. Este estudio tuvo por mira correlacionar la evolución clínica y los electrocardiogramas obtenidos antes y después de operaciones torácicas en 112 enfermos.

2. En 30 por ciento de toda la serie de enfermos descubriéronse modificaciones en las Derivaciones I y IV de la onda en T, en forma de inversión de la T_1, ya sola o unida a inversión de la T_4, en ausencia completa de toda cardiopatía observable.

3. Ofrécense argumentos para confutar la suposición de la coexistencia de alteraciones miocardíacas en estos enfermos.

4. Abógase por que continúen las investigaciones de las alteraciones descritas.

Acknowledgments

The writers are indebted to J. D. Murphy, M. D., Chief of the Surgical Service, and his staff for their indispensable services in making this study possible, and to Mrs. Mildred Knowlton, Medical Librarian, for valuable assistance in translation of foreign articles and preparation of the final manuscript.

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STREPTOMYCIN-TUBERCULOSIS RESEARCH PROJECT OF THE AMERICAN TRUDEAU SOCIETY[1,2,3,4]

A Summary Report

H. McLEOD RIGGINS[5] AND H. CORWIN HINSHAW[5]

INTRODUCTION

In the fall of 1946, after it had been demonstrated (1, 2, 3, 4, 8, 9) that streptomycin had definite therapeutic potentialities in the treatment of clinical tuberculosis, the manufacturers of streptomycin donated a large quantity to the American Trudeau Society for expansion of clinical research. The primary purpose of the investigation was to explore the integration of streptomycin therapy with other therapeutic procedures in the treatment of *pulmonary* types of the disease. Although much had already been learned concerning the limitations and therapeutic potentialities of streptomycin in the treatment of tuberculosis, nevertheless there still remained many questions only partially answered. Some of the major problems were: (*1*) determination of the types of pulmonary tuberculosis most likely to benefit from the use of streptomycin; (*2*) the most satisfactory methods and frequency of treatment; (*3*) the optimum daily dosage; (*4*) the optimum duration of treatment; (*5*) the most timely integration of streptomycin with other therapeutic procedures, especially collapse therapy and lung resection; (*6*) the determination of the incidence and clinical significance of the emergence of strains of tubercle bacilli less sensitive to the drug and how this phenomenon might be delayed, minimized or prevented; (*7*) the incidence, severity, clinical significance and, if possible, the prevention of serious toxic manifestations, particularly deafness and vestibular dysfunction; and (*8*) relapse of the disease.

The Executive Committee of the American Trudeau Society designated a group of clinical and laboratory investigators, who later functioned as Clinical and Laboratory Chemotherapy Subcommittees of the Committee on Medical Research and Therapy, to collaborate in carrying out studies on the above and other problems. Members of the Clinical Chemotherapy Subcommittee were: Dr. J. Burns Amberson, New York, New

[1] The full report, including chapters by those participating in the project and by others on certain fundamental aspects of the problem, is now being compiled and edited for publication in book form by the National Tuberculosis Association.

[2] For previous report refer to H. McLeod Riggins and H. Corwin Hinshaw, The Streptomycin-Tuberculosis Research Project, American Trudeau Society, Am. Rev. Tuberc., 1947, *56*, 168.

[3] This study was aided by grants from the Division of Research Grants and Fellowships of the National Institute of Health, U. S. Public Health Service and from the Medical Research Committee of the National Tuberculosis Association.

[4] The streptomycin was generously donated by Abbott Laboratories, Eli Lilly and Co., Merck and Co., Inc., Chas. Pfizer and Co., E. R. Squibb & Sons, and The Upjohn Company.

[5] Present (1947–1948) and previous (1946–1947) Chairmen of the Subcommittee on Chemotherapy, Committee on Medical Research and Therapy, American Trudeau Society.

York; Dr. Emil Bogen, Olive View, California; Dr. H. Corwin Hinshaw, Rochester, Minnesota (Chairman 1946–1947); Dr. Kirby S. Howlett, Jr., Shelton, Connecticut; Dr. Walsh McDermott, New York, New York; Dr. Edward N. Packard, Trudeau, New York; Dr. H. McLeod Riggins, New York, New York (Chairman 1947–1948); and Dr. H. Stuart Willis, McCain, North Carolina.

Members of the Laboratory Chemotherapy Subcommittee were: Dr. Emil Bogen, Olive View, California; Dr. H. Corwin Hinshaw, Rochester, Minnesota; Mr. William Steenken, Jr., Trudeau, New York; Dr. H. Stuart Willis, McCain, North Carolina; Dr. C. Eugene Woodruff, Northville, Michigan; and Dr. Guy P. Youmans, Chicago, Illinois (Chairman).

In order to coordinate the laboratory studies in connection with the project and to ensure more adequate facilities and uniform procedures, three district laboratories were designated at: Olive View Sanatorium, Olive View, California, Dr. Emil Bogen, Director; Trudeau Sanatorium, Trudeau, New York, Mr. William Steenken, Jr., Director; and Northwestern University Medical School, Chicago, Illinois, Dr. Guy P. Youmans, Director.

The present communication is in no way to be construed as a comprehensive report covering the studies of the individual investigators, but rather as a report in summary form including discussions and tentative opinions with regard to some of the more important aspects of the study. A comprehensive report, including chapters by the individual investigators and more extensive tabular and graphic data, will be published in book form at an early date by the National Tuberculosis Association.

OBSERVATIONS

Clinical Material

A total of 566 patients were studied between September, 1946 and January, 1948. Information on these patients was obtained from the eight investigators through the medium of questionnaires. A comprehensive statistical analysis was carried out on the pertinent findings in 332 patients with pulmonary tuberculosis who had completed streptomycin therapy and had been observed for at least 90 days after the beginning of treatment. Reports on the remaining 234 patients are not included in the present analysis because they were either treated for less than 30 days, had been observed for less than 90 days after the beginning of treatment (at the time the questionnaires were returned), had received less than 0.5 Gm. of streptomycin daily, or were patients with meningeal or miliary tuberculosis or had some type of extrapulmonary tuberculosis only.

Of the 332 patients with pulmonary tuberculosis reported on at this time, 17 had minimal, 118 moderately advanced and 197 far advanced disease. Eighty per cent of these patients were stated by the investigators to have predominantly acute or subacute disease, and the remainder predominantly chronic disease at the beginning of streptomycin treatment (figure 1).

One hundred seventy-seven of these 332 patients with pulmonary tuberculosis had a total of 204 known tuberculous complications. The principal complications were: (*1*) tracheobronchial and laryngeal tuberculosis, 125; (*2*) lymph

node and sinus tract tuberculosis, 18; (*3*) intestinal tract tuberculosis, 16; (*4*) bone and joint tuberculosis, 13; (*5*) genito-urinary tract tuberculosis, 10; and (*6*) miscellaneous complications, 22.

Grouping of the 332 patients according to race, sex, and age showed the following: white, 262; Negro, 36; other, 34; female, 200; male, 132. Sixty-six per cent of the patients were between 20 and 39 years of age.

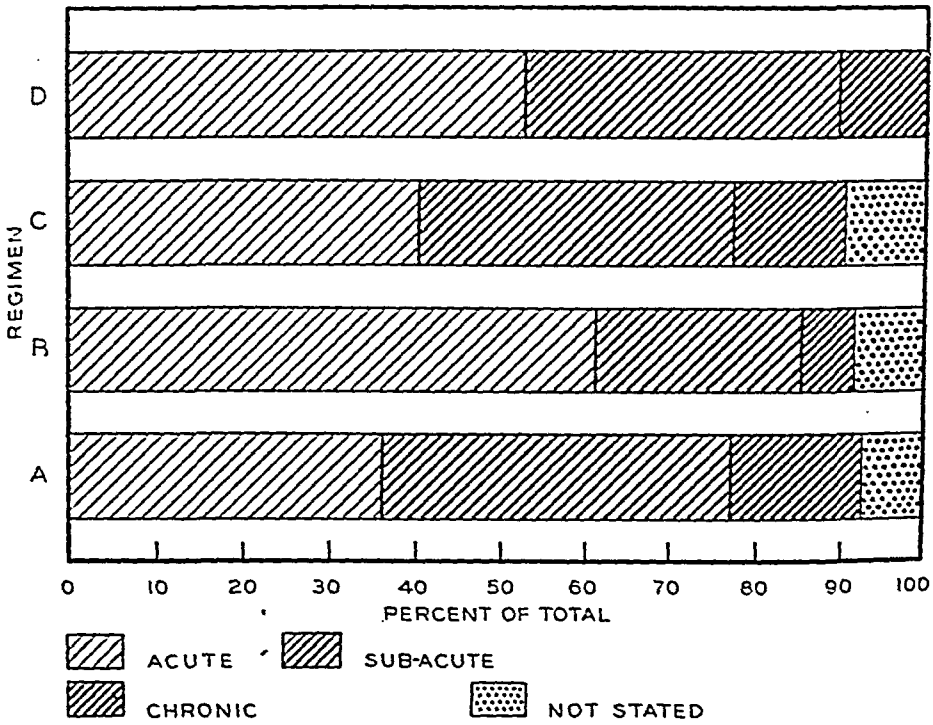

FIG. 1. Clinical types of disease, at beginning of streptomycin therapy, in 332 patients with pulmonary tuberculosis (according to regimen).

Regimen	*Duration of Treatment* (days)	*Daily Dosage Streptomycin* (grams)
A	30 to 89	0.5 to 1.4
B	30 to 89	1.5 to 3.0
C	90 or over	0.5 to 1.4
D	90 or over	1.5 to 3.0

Relationship of Streptomycin Efficacy to the Type and Extent of Pulmonary Pathology

Before considering the comparative therapeutic advantages and limitations of different streptomycin regimens, consideration should be given to the more important basic factors which significantly influence or largely determine the therapeutic potentialities of any antibacterial drug in tuberculosis. These factors are the character and extent of the predominant tuberculous disease, the resulting physiologic abnormalities, the immediate and remote potentialities of the disease,

the native and acquired resistance of the patient, and other factors which may significantly limit the successful application of a comprehensive and properly integrated therapeutic program. The mechanism of action of streptomycin in inhibiting the disease process affords additional insight into the potentialities and limitations of streptomycin.

The beneficial effect of streptomycin in tuberculosis is dependent upon its interference with the growth and multiplication of streptomycin-sensitive strains of *M. tuberculosis* (5). Streptomycin apparently does not directly enhance tissue repair but it probably does favorably influence the tempo and quality of repair by inhibiting the growth and multiplication of tubercle bacilli. Assuming this to be true, it would be expected that the response of the tuberculous lesion to streptomycin would be fundamentally related to the characteristics of the pathology and the patient's ability to cope with his disease. It has been shown, experimentally and clinically, that streptomycin is more effective against relatively early acute and subacute lesions, in which the extent and degree of caseation is limited, than it is against confluent and rapidly caseating and liquefying tuberculous pneumonias, or against extensive chronic fibrocaseous and cavernous disease. Under certain circumstances, therefore, streptomycin appears to bring into favorable balance resistance and repair over susceptibility and destruction. In view of the mechanism by which streptomycin appears to affect certain tuberculous lesions beneficially, it cannot be expected that the drug will exert a direct favorable influence on such irreversible pathological lesions as extensive caseation and fibrosis. It may logically be expected, however, that streptomycin can favorably influence the behavior of some pulmonary cavities, especially those which tend to increase in size as the result of progressive inflammation, caseation, and liquefaction, or certain of those whose size and behavior are influenced by acute and ulcerative tuberculous bronchitis. If the behavior of the pulmonary cavity is related to an active tuberculous bronchitis, healing of the bronchial disease and the re-establishment of normal bronchial function, as a result of streptomycin therapy, may significantly aid closure and healing of the cavity.

These fundamental factors, often minimized or overlooked entirely, play a more decisive role in the degree of therapeutic effectiveness of streptomycin and the ultimate outcome of the issue than do the often overemphasized differences in total daily dosage or duration of treatment, as important as these latter factors are. In this connection, it should be stressed that in advanced progressive chronic tuberculosis the present clinical and roentgenologic methods for determining the different types of pathological processes in their true proportions, and classifying them, are relatively crude and inaccurate at best. The effect of streptomycin has frequently emphasized the limitations of these methods. Frequently, disease presumed to be predominantly caseous and at other times predominantly productive undergoes marked resolution and clearing and, perhaps equally often, disease presumed to be largely inflammatory in nature proves to be caseous. The practical difficulties, therefore, of choosing essentially comparable types of disease, particularly from the point of view of the effect of a chemotherapeutic agent, in equal proportions in different regimens become clear.

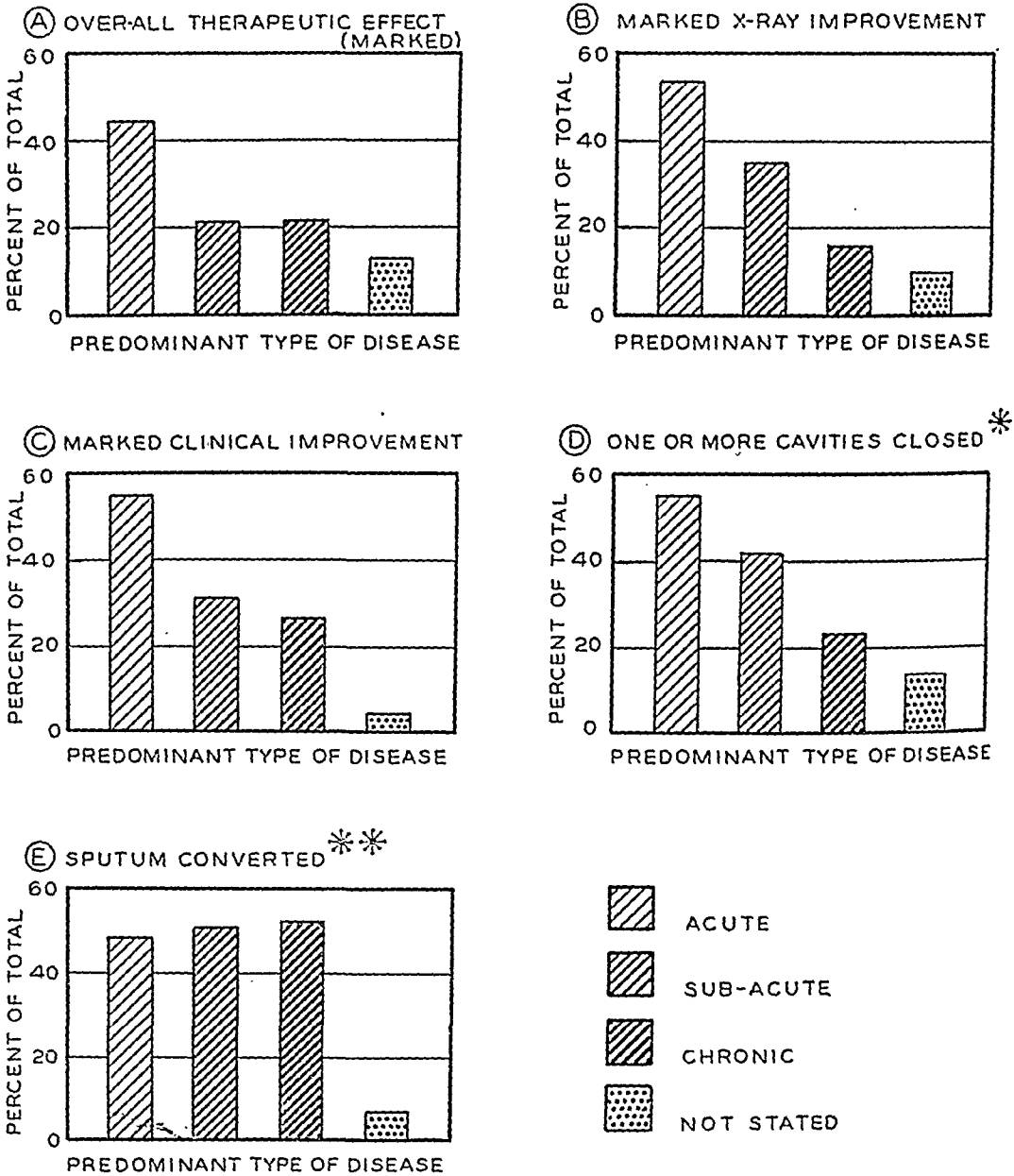

FIG. 2. Individual investigators' estimate of the efficacy of streptomycin, in relation to the predominant type of disease, in 332 patients with pulmonary tuberculosis, observed from six to ten months after the beginning of treatment.

* Percentages based upon 218 patients with demonstrable cavities at the beginning of streptomycin treatment.

** Percentages based upon 284 patients with sputum or gastric lavage contents positive for tubercle bacilli.

It is equally obvious that erroneous conclusions may be drawn if the differences in the results obtained are solely attributed to the differences in regimens used. This is especially true if small numbers of patients are involved.

An attempt was made to evaluate the therapeutic effect of streptomycin on the different clinical types of disease in 332 patients with pulmonary tuberculosis classified by the investigators as predominantly acute or subacute and predominantly chronic. The results are shown in figure 2. The effectiveness of the antibiotic on the three types of disease was measured in terms of: (*1*) clinical improvement, (*2*) roentgenographic improvement, (*3*) closure of cavities, (*4*) "conversion of sputum," and (*5*) over-all marked improvement. In general, the therapeutic effectiveness of streptomycin was strikingly greater in patients with recent and presumably acute exudative disease. The drug also appeared to be more effective against subacute than against chronic disease. The degree of its effectiveness against subacute disease, however, was not as consistent or striking as against the more recent and acute exudative disease. The least beneficial effects were noted in patients with extensive predominantly chronic disease.

One of the significant limitations of streptomycin, with regard to all three types of disease, and irrespective of the regimen used, was emphasized by the similarity of the sputum findings. There was no significant difference found in "sputum conversion" in the different types of disease.

As the degree of effectiveness of streptomycin is fundamentally related to the type and extent of pathology, and as it appears to be least effective against extensive chronic disease, it should not be anticipated that streptomycin and bed-rest alone would bring about arrest of the disease in the great majority of patients with advanced chronic cavitary tuberculosis.

Streptomycin Regimens

None of the investigators was restricted to the exploration of a single regimen and as a rule each investigator used several different regimens. A few investigators explored many regimens. Because of the multiplicity of regimens employed it was impossible to evaluate all of them individually. Therefore, for statistical comparison of four basic regimens, the 332 patients with pulmonary tuberculosis who were observed for at least ninety days after the beginning of treatment were arbitrarily divided into four groups.

Regimen A. 124 patients	Patients treated for 30 to 89 days, with a daily dosage of streptomycin varying from 0.5 Gm. to 1.4 Gm.
Regimen B. 46 patients	Patients treated for 30 to 89 days, with a daily dosage of streptomycin varying from 1.5 Gm. to 3.0 Gm.
Regimen C. 89 patients	Patients treated for 90 days or over, with a daily dosage of streptomycin varying from 0.5 Gm. to 1.4 Gm.
Regimen D. 73 patients	Patients treated for 90 days or over, with a daily dosage of streptomycin varying from 1.5 Gm. to 3.0 Gm.

The most important facts brought out in table 1 are: (*1*) Of the 213 patients receiving daily dosages varying from 0.5 Gm. to 1.4 Gm., 148 (70 per cent) received 1.0 Gm. daily. (*2*) Of the 119 patients receiving daily dosages varying from 1.5 Gm. to 3.0 Gm., only 10 (8 per cent) received 3.0 Gm. daily. In the evaluation of results according to regimens it is important to bear in mind the fact that a great majority of the patients in regimens A and C received a daily dosage of 1.0 Gm. and in regimens B and D only a few patients received 3.0 Gm. daily.

TABLE 1

Breakdown of total patients in each regimen according to average daily dosage

AVERAGE DAILY DOSAGES	REGIMEN A NUMBER OF PATIENTS	REGIMEN B NUMBER OF PATIENTS	REGIMEN C NUMBER OF PATIENTS	REGIMEN D NUMBER OF PATIENTS
0.5 Gm.	12	—	8	—
0.6 to 0.9 Gm.	12	—	10	—
1.0 Gm.	94	—	54	—
1.1 to 1.4 Gm.	6	—	17	—
1.5 to 1.9 Gm.	—	13	—	34
2.0 to 2.9 Gm.	—	24	—	38
3.0 Gm.	—	9	—	1
Total	124	46	89	73

To evaluate different regimens providing different durations of treatment with accuracy, it is essential to have the results analyzed at the end of *comparable* periods of time after the *beginning* of streptomycin therapy. In this study, results obtained in the treatment of patients with regimen A were analyzed an average of 182 days after the beginning of treatment; with regimen B, an average of 243 days after the beginning of treatment; with regimen C, an average of 264 days after the beginning of treatment; and with regimen D, an average of 294 days after the beginning of treatment.

Any suggestions, trends, or tentative opinions gained from such an analysis as this must of necessity be considered in the light of the inherent and obvious deficiencies of this and similar cooperative studies involving a number of investigators. This project does have certain qualities to recommend it, however, a few of which might be noted. The investigation represents the combined thinking and efforts of a coordinated national group of clinical and laboratory investigators associated with representative medical schools and tuberculosis teaching centers. The clinical material consists of a wide sampling of pulmonary tuberculosis from the point of view of the different types of disease, age, race, and sex of the patients treated in widely separated sections of the country. All information and opinions concerning individual patients, including evaluation of results, are entirely those of the individual investigators. The compiling and tabulating of the statistical material were carried out by Dr. Carroll E. Palmer and Mrs. Shirley H. Ferebee and their staff of the U. S. Public Health Service. None of this group had an official connection with the streptomycin study of the American Trudeau Society. Finally, the number of patients observed was considerably larger than could have been studied by a single investigator at the time and is considered sufficiently large to be of statistical significance.

Evaluation of Streptomycin Regimens

Evaluation of the results obtained on the four regimens is based upon the reports of the individual investigators concerning clinical and roentgenographic

findings, closure of cavities, "conversion of sputum," estimates of the over-all therapeutic effect of the drug, toxic manifestations of streptomycin, emergence of strains of tubercle bacilli less sensitive to streptomycin, and relapse of the disease. As the results could have been partially affected by such other therapeutic procedures as bed-rest and collapse therapy, the value of streptomycin was estimated in each instance by the physician treating the patient.

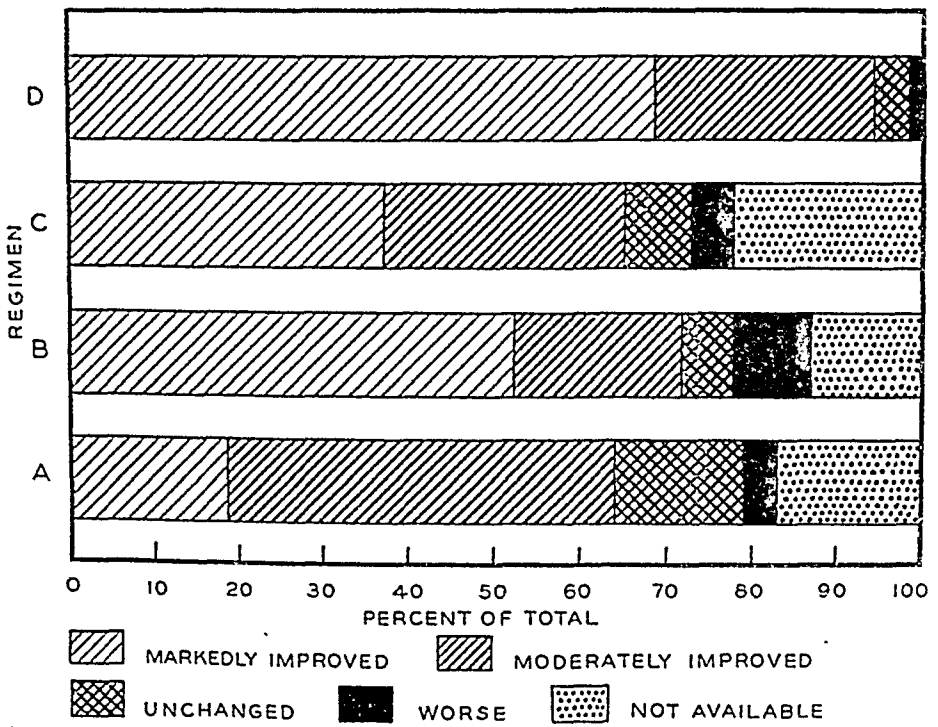

FIG. 3. Clinical condition, at most recent examination, of 332 patients with pulmonary tuberculosis observed from six to ten months after start of streptomycin, in comparison with beginning of treatment (according to regimen).

Regimen	*Duration of Treatment* (days)	*Daily Dosage Streptomycin* (grams)
A	30 to 89	0.5 to 1.4
B	30 to 89	1.5 to 3.0
C	90 or over	0.5 to 1.4
D	90 or over	1.5 to 3.0

Daily Dosage and Duration of Treatment

Despite the fundamental factors which influence the effectiveness of streptomycin, a comparison of the results obtained on the four different regimens appears to indicate some relationship between the size of the daily dosage and the therapeutic results attained. Moreover, a similar though less definite relationship appears to exist between the duration of treatment and the therapeutic results. It is to be re-emphasized, however, that the relationship which appears to exist

between therapeutic results and daily dosage may be more *apparent* than *real*, because of the difference in the percentage distribution of the clinical types of cases treated in the four regimens (figure 1) and for the other reasons previously mentioned.

Clinical improvement: As may be seen in figure 3, clinical improvement occurred in 86 per cent of the patients receiving the larger daily dosages (1.5 Gm. to 3.0 Gm., regimens B and D) in contrast to 65 per cent of the patients who

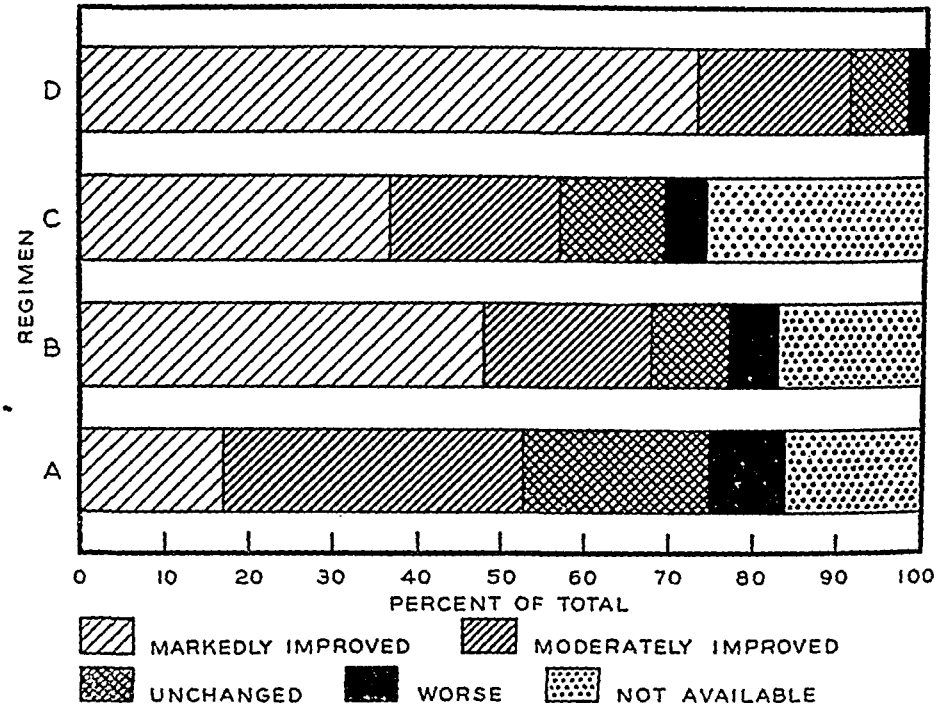

FIG. 4. Condition, according to most recent roentgenological findings, of 332 patients with pulmonary tuberculosis observed from six to ten months after start of streptomycin, in comparison with beginning of treatment (according to regimen).

Regimen	*Duration of Treatment* (days)	*Daily Dosage Streptomycin* (grams)
A	30 to 89	0.5 to 1.4
B	30 to 89	1.5 to 3.0
C	90 or over	0.5 to 1.4
D	90 or over	1.5 to 3.0

received the smaller daily dosages (0.5 Gm. to 1.4 Gm., regimens A and C). Clinical improvement seems related *much less* to duration of treatment, in so far as a comparison of the two periods of treatment used (30 to 89 days, and 90 days or over) is concerned. In the patients receiving treatment for the longer periods of time (90 days or over, regimens C and D) 78 per cent showed clinical improvement, while in the patients receiving treatment for the shorter periods of time (30 to 89 days, regimens A and B) 66 per cent showed clinical improvement.

Roentgenographic improvement: The frequency and degree of the over-all roentgenographic improvement seem to be related to the total daily dosage (figure 4). This is shown by comparing the results obtained on regimens A and B, C and B, and C and D. In each instance it will be seen that the best results were obtained in patients receiving the larger daily dosages. Comparatively better results were obtained on regimen B than on regimen C, even though pa-

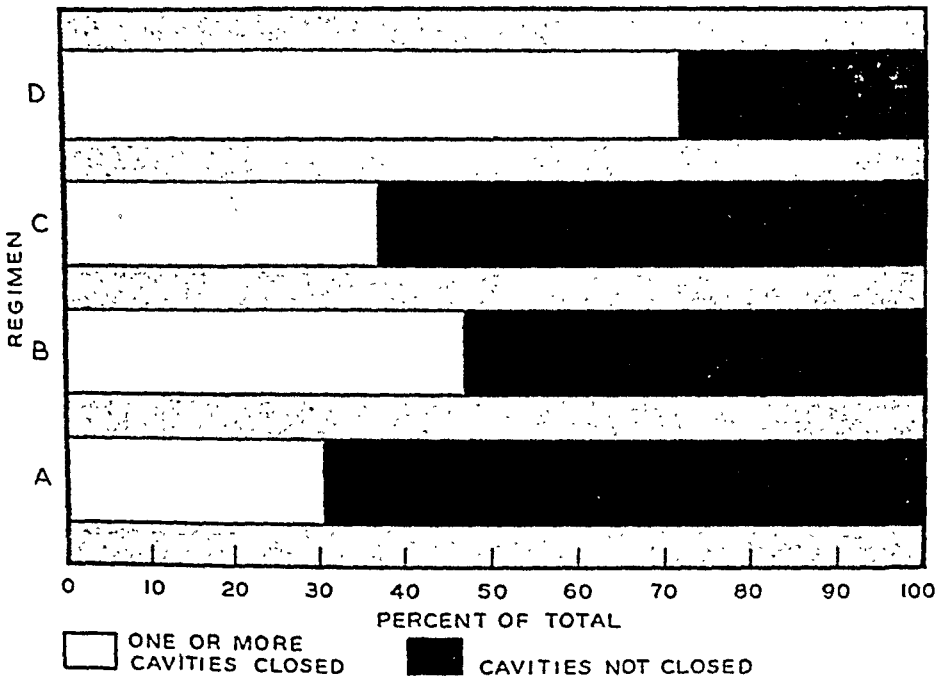

FIG. 5. Closure of one or more cavities, during and after streptomycin therapy, in 218 patients with pulmonary tuberculosis observed from six to ten months after the beginning of treatment (according to regimen).

(Only 218 of the 332 patients had demonstrable cavities at the beginning of streptomycin therapy.)

Regimen	*Duration of Treatment* (days)	*Daily Dosage Streptomycin* (grams)
A	30 to 89	0.5 to 1.4
B	30 to 89	1.5 to 3.0
C	90 or over	0.5 to 1.4
D	90 or over	1.5 to 3.0

tients on the latter regimen were treated for a longer period of time. Roentgenographic improvement was noted in 82 per cent of the patients receiving the larger daily dosages (regimens B and D) and in 54 per cent of the patients receiving the smaller daily dosages (regimens A and C). The over-all roentgenographic improvement seems also to be related, but to a *much less* extent, to the duration of treatment. This is shown by comparing the combined results obtained on regimens A and B with the combined results on regimens C and D. Roentgeno-

graphic improvement was noted in 72 per cent of the patients receiving treatment for the longer periods of time (90 days or over, regimens C and D), and in 56 per cent of the patients who received treatment for the shorter periods of time (30 to 89 days, regimens A and B).

Closure of cavities: In general, the study revealed that the same relationship appears to exist between the closure of cavities and the total daily dosage and

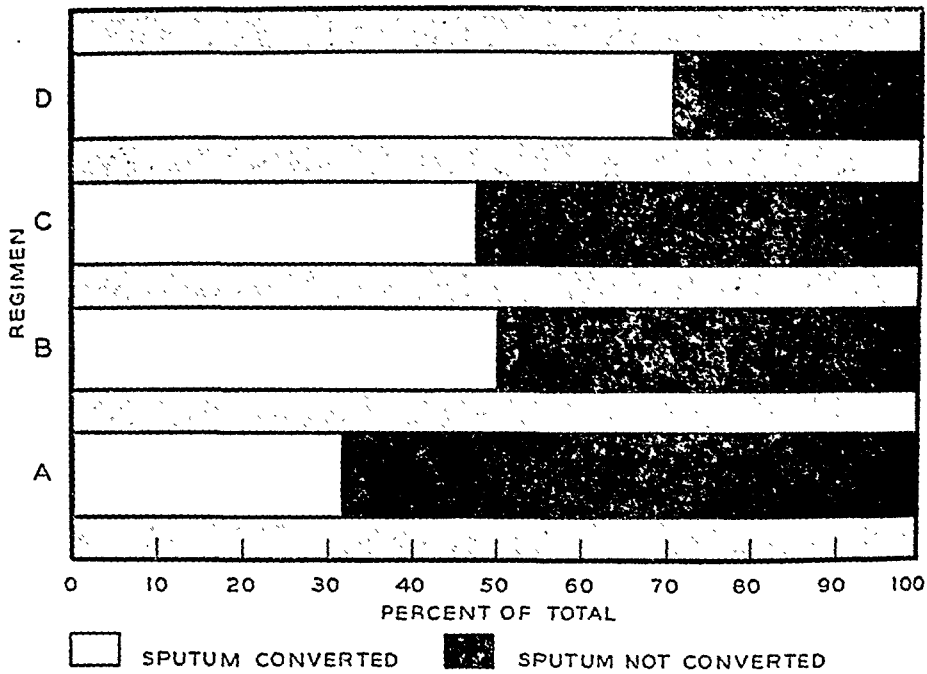

FIG. 6. Conversion of sputum from positive to negative for tubercle bacilli, during or after streptomycin therapy, in 284 patients with pulmonary tuberculosis observed from six to ten months after the beginning of treatment (according to regimen).

(The remaining 48 patients had recently had sputum or gastric lavage contents positive for tubercle bacilli.)

Regimen	*Duration o Treatment* (days)	*Daily Dosage Streptomycin* (grams)
A	30 to 89	0.5 to 1.4
B	30 to 89	1.5 to 3.0
C	90 or over	0.5 to 1.4
D	90 or over	1.5 to 3.0

duration of treatment as was found in analysis of the over-all roentgenographic and clinical improvement (figure 5). That is to say, the closure of one or more cavities occurred in 62 per cent of those patients who received the larger daily dosages of streptomycin in contrast to 34 per cent in the patients receiving the smaller daily dosages. Closure of one or more cavities occurred in 51 per cent of patients receiving treatment for the longer periods of time (90 days or over, regimens C and D), while it occurred in 36 per cent of the patients receiving

treatment for the shorter periods of time (30 to 89 days, regimens A and B). Follow-up of some of these patients, however, has shown that cavities which disappeared on the chest roentgenogram or were apparently closed have reopened. Such findings provide support for the opinion that streptomycin is usually not a definitive form of therapy, especially for the more destructive and chronic types of tuberculosis. If durable results are to be obtained, it is always

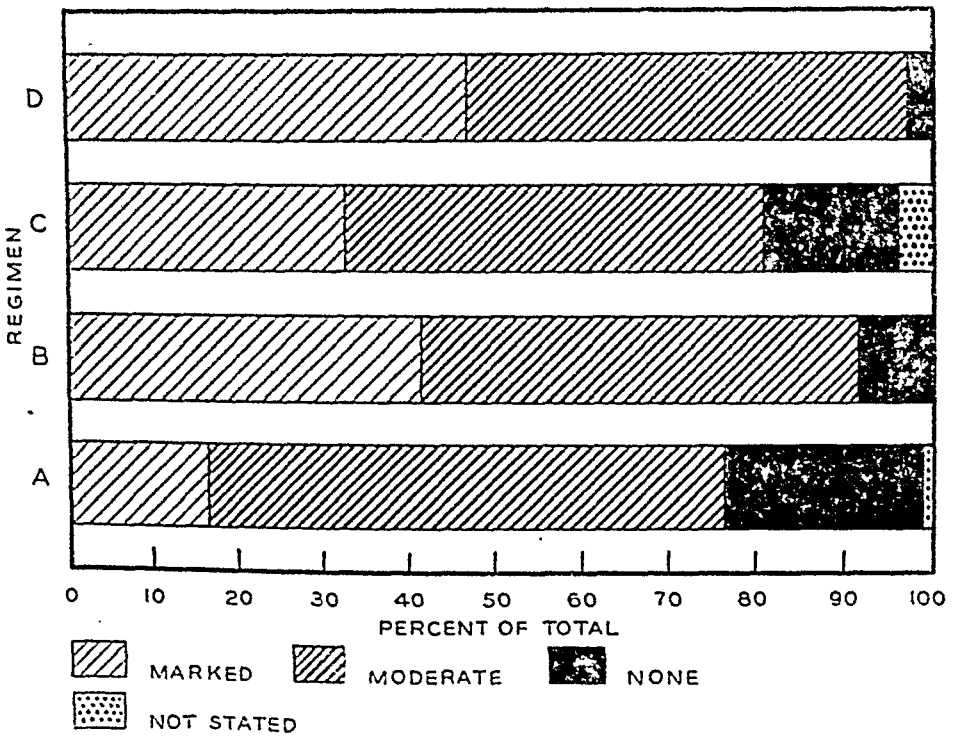

FIG. 7. Individual investigators' estimate of the over-all efficacy of streptomycin in 332 patients with pulmonary tuberculosis, observed from six to ten months after the beginning of treatment (according to regimen).

Regimen	*Duration of Treatment* (days)	*Daily Dosage Streptomycin* (grams)
A	30 to 89	0.5 to 1.4
B	30 to 89	1.5 to 3.0
C	90 or over	0.5 to 1.4
D	90 or over	1.5 to 3.0

essential to supplement streptomycin with bed-rest and frequently with some form of collapse therapy or occasionally lung resection.

"Conversion of sputum": The disappearance of tubercle bacilli from the sputum occurred more frequently in the patients receiving the larger daily dosages of streptomycin (63 per cent) than in the patients receiving the smaller daily dosages (38 per cent). Reversal of infectiousness also appears to be related to the duration of treatment. In the patients receiving treatment for the longer periods

of time (90 days or over) "sputum conversion" occurred in 58 per cent, while in the patients treated for the shorter periods of time (30 to 89 days) "conversion" occurred in 36 per cent.

The unusually high percentage of "sputum conversions" in all regimens may be partially explained by the fact that "conversion" of sputum in this series of patients was based upon the results of the last sputum examination and in many instances the type of examination was by concentration method. The high percentages of "sputum conversion" would probably be much lower if they were based upon repeated examination by culture or animal inoculation.

The follow-up of certain patients whose sputum was "converted" shows that not infrequently the sputum again becomes positive for tubercle bacilli. It is believed that the increment of reopened cavities and the return of infectiousness may increase significantly with the passage of time following completion of streptomycin therapy, particularly if other therapeutic procedures are not properly applied at the opportune time. It should be borne in mind that the results were evaluated from 182 to 294 days after the beginning of streptomycin treatment.

The consistency of the relationship between the total daily dosage and therapeutic results and, to a less degree, between the duration of treatment and therapeutic results is still further suggested by the reports of the individual investigators of the estimated over-all therapeutic effects of streptomycin obtained on the four regimens (figure 7).

Sensitivity of Tubercle Bacilli to Streptomycin before, during and after Completion of Therapy

It has been well established by many workers that human strains of *M. tuberculosis* which have not been previously exposed to streptomycin are sensitive *in vitro* to low concentrations of the antibiotic. Studies have also shown that a high percentage of strains of tubercle bacilli become less sensitive to streptomycin in proportion to the *duration* of exposure to the drug. It has occasionally been noted that organisms still sensitive to streptomycin at the completion of treatment may become less sensitive at some later date and that the degree of "drug fastness" present at the end of treatment may actually increase following cessation of treatment. It is important, therefore, to determine the sensitivity of the organisms routinely before retreatment is undertaken.

The possibility of the existence of a relationship between the daily dosage of streptomycin and the rapidity and frequency of the emergence of significantly resistant strains during treatment (10 or more γ of streptomycin per ml.) has potential implications concerning optimum daily dosage. The few data available regarding this possible relationship will be presented. It is also possible that the simultaneous use of certain tuberculostatic agents with streptomycin may perhaps delay or decrease the emergence of streptomycin-resistant organisms. Should further studies along this line prove encouraging, large scale trials of combined chemotherapy would seem definitely warranted for the purpose of clarifying this question. A few such studies are already under way.

A limited amount of information concerning the emergence of less sensitive

strains of tubercle bacilli during streptomycin therapy is available from this study. As there is increasing evidence that *in vitro* resistance is indicative also of *in vivo* resistance and that microorganism resistance of 10 or more γ of streptomycin per ml. is of some clinical significance, data will be presented only on those patients whose tubercle bacilli became resistant to 10 or more γ of streptomycin. Reliable data concerning microorganism sensitivity during streptomycin treatment are available on 159 of the 332 patients in the series. Data on the remaining patients are incomplete for various reasons but largely because bacilli were not available for testing at the completion of treatment.

Of the 159 patients, 56 (35.2 per cent) are known to have discharged tubercle bacilli resistant to 10 or more γ of streptomycin per ml. at various periods of time after the beginning of treatment. Of 17 patients whose bacilli were studied for streptomycin sensitivity one to 30 days after the start of treatment, 2 (11.7 per cent) had organisms resistant to 10 or more γ per ml.; of 40 patients whose organisms were studied 31 to 60 days from the start of treatment, 12 (30 per cent) had organisms resistant to 10 or more γ per ml.; of 27 patients whose bacilli were studied 61 to 90 days after the start of treatment, 12 (44.4 per cent) had organisms resistant to 10 or more γ per ml.; of 23 patients whose bacilli were studied 91 to 120 days after the start of treatment, 12 (52.2 per cent) had organisms resistant to 10 or more γ per ml.; and of 52 patients whose bacilli were studied 121 days or later after the start of treatment, 18 (34.6 per cent) had organisms resistant to 10 or more γ per ml. In the remaining 103 patients (64.9 per cent) studied periodically during part of the treatment period, the sputum either became negative or the organisms remained sensitive to less than 10 γ of streptomycin per ml. It is believed that, if it were possible to determine the streptomycin sensitivity of tubercle bacilli in all patients at the end of prolonged treatment, there would be a much greater incidence of significantly resistant strains than indicated above.

These data are too few to permit a precise analysis of the relation of the length of treatment to either the incidence or the time of appearance of drug-resistant strains of tubercle bacilli. Nevertheless, examination of the data does reveal that the emergence of what appear to be significantly resistant strains (resistant to 10 or more γ streptomycin per ml.) did occur in a fairly high percentage (35.2 per cent) of the patients.

Possibly of equal importance is the fact that there appears to be some relationship between the total daily dosage and the rapidity of emergence of organisms resistant to at least 10 γ of streptomycin per ml. Although the number of patients in this group is too small to have statistical significance, nevertheless it is of interest to note that the emergence of organisms resistant to 10 γ and over per ml. was more rapid in patients receiving the *larger* daily dosages of streptomycin (1.5 Gm. to 3.0 Gm., regimens B and D) than in patients receiving the smaller daily dosages (0.5 Gm. to 1.4 Gm., regimens A and C) (figure 8). It is probably of equal importance, however, to note that a *greater* percentage of patients who received the *smaller* daily dosages showed emergence of strains resistant to 10 or more γ per ml. Of the 85 patients receiving the *smaller* daily

dosages, 35 (41.2 per cent) had organisms resistant to 10 or more γ per ml. at different periods of time after the start of treatment, and of the 74 patients receiving the larger daily dosages, only 21 (28.4 per cent) had organisms resistant to 10 or more γ per ml. after beginning of treatment.

These data would seem to pose another question with regard to optimum daily dosage. Considered only in the light of the rapidity of emergence of organisms significantly resistant to streptomycin, it might be assumed, if these findings

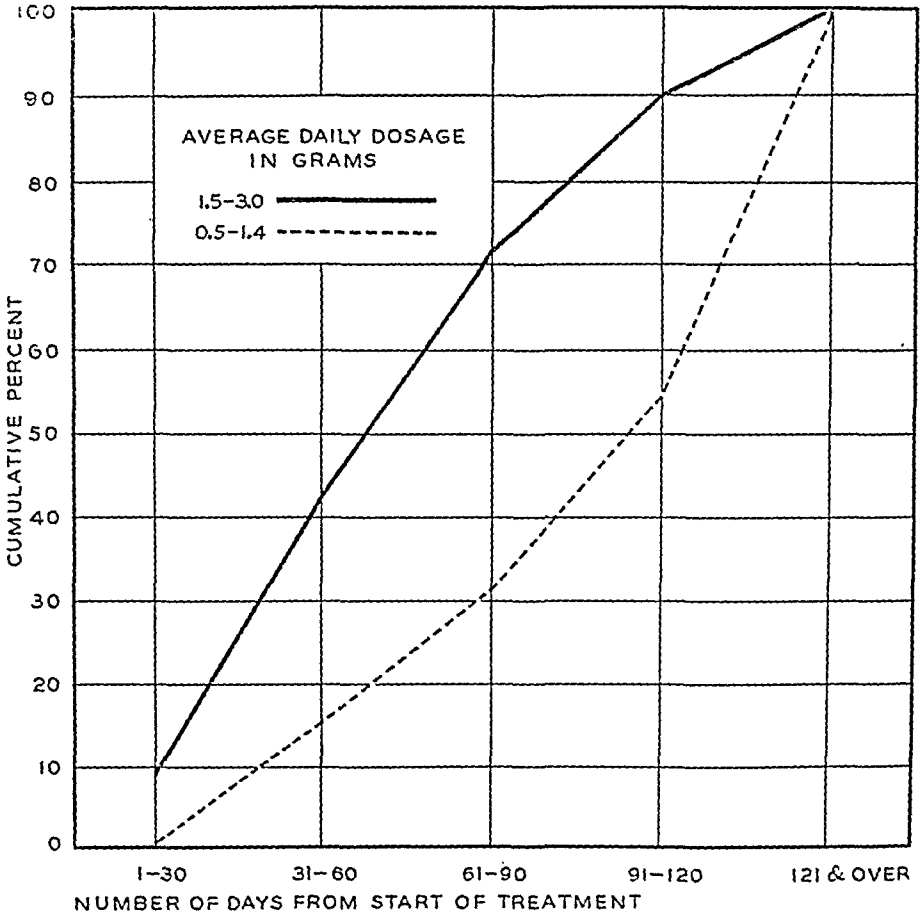

FIG. 8. Emergence of strains of tubercle bacilli resistant to 10 or more γ of streptomycin per ml., in relation to duration of treatment and average daily dosage.

are confirmed, that an advantage of smaller daily dosages is that they presumably cause the emergence of significantly resistant organisms less rapidly than do larger daily dosages. For the group of 56 patients whose microorganisms were resistant to 10 or more γ of streptomycin per ml., the monthly percentages[6] and the cumulative percentage of emergence of this degree of drug-resistance are shown in figure 8.

[6] Sensitivity tests were done periodically from start of treatment.

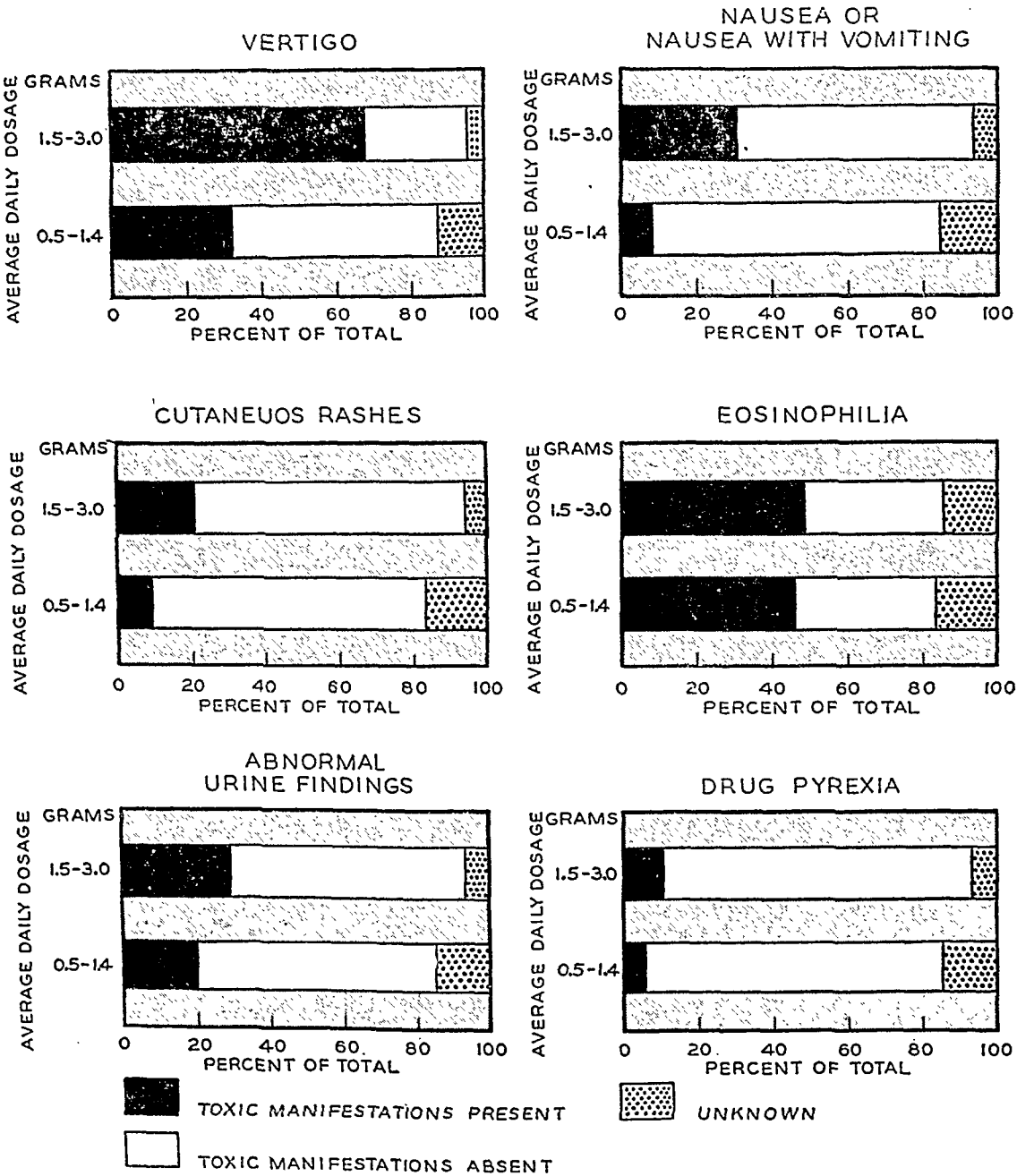

FIG. 9. Toxic manifestations encountered in 332 patients with pulmonary tuberculosis, according to average daily dosages of streptomycin.

Toxic manifestations: The frequency and severity of toxic manifestations were consistently *greater* in the patients receiving the *larger* daily dosages of streptomycin (1.5 Gm. to 3.0 Gm., regimens B and D) than in the patients receiving the smaller daily dosages (0.5 Gm. to 1.4 Gm., regimens A and C) (figure

9). Named in the order of their importance and frequency, the toxic manifestations encountered in this series were: vestibular dysfunction; eosinophilia; nausea and vomiting; evidence of renal irritation or damage which was usually transient (severe renal damage due primarily to streptomycin was not encountered); dermatitis; and drug pyrexia. In the great majority of instances toxic manifestations were reversible, labyrinthine dysfunction usually being either reversible or compensated for in almost all cases. Permanent deafness due to streptomycin

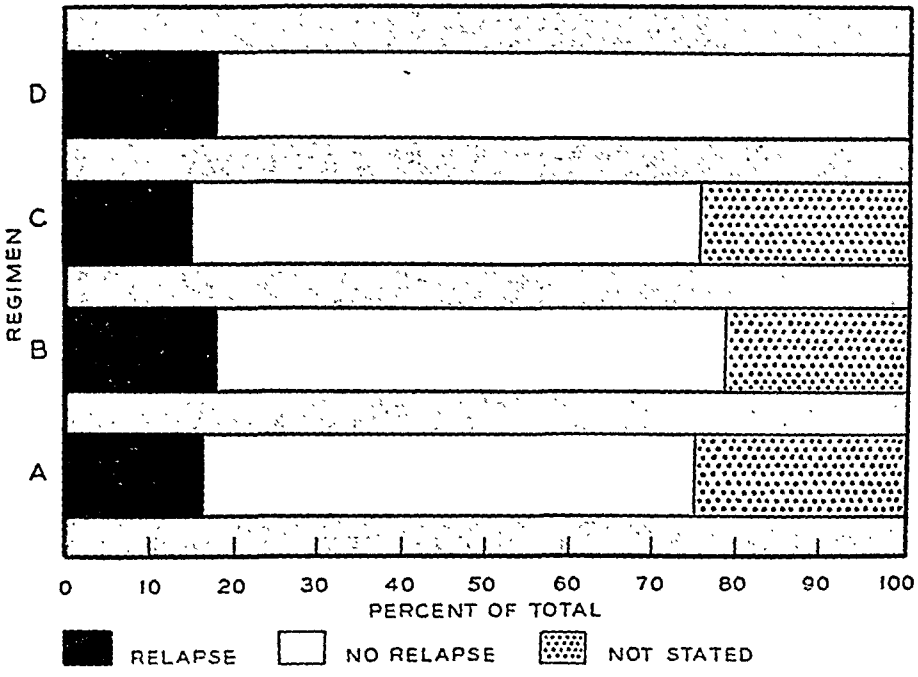

FIG. 10. Relapse of the disease, during or after streptomycin therapy, in 332 patients with pulmonary tuberculosis observed from six to ten months after the beginning of treatment (according to regimen).

Regimen	*Duration of Treatment* (days)	*Daily Dosage Streptomycin* (grams)
A	30 to 89	0.5 to 1.4
B	30 to 89	1.5 to 3.0
C	90 or over	0.5 to 1.4
D	90 or over	1.5 to 3.0

was not reported by the investigators in this series of pulmonary tuberculosis patients.

In the last analysis, determination of the optimum total daily dosage is based upon the possible degree of therapeutic effectiveness, on the one hand, and the possible severity of the toxic effects, on the other. Within certain limits, the selection of the optimum daily dosage is related to the threat of the disease versus the potential untoward toxic effects. For example, it might be pointed out that it is better to use a relatively large dosage of streptomycin and cure a

tuberculous meningitis, even though deafness supervenes, than to use, in attempting to avoid toxic effects, ineffective dosage and lose the patient. The same reasoning does not always apply, however, with regard to the use of larger daily dosage (20 to 40 mg. per kilogram of body weight) for many patients with pulmonary tuberculosis. In these patients the threat of the disease to life is usually not as definite and immediate, nor is streptomycin therapy definitive but must usually be accompanied by other therapeutic procedures if durable results are to be achieved. It is strongly believed that the optimum daily dosage cannot be generalized but must be individualized, as in other drug therapy.

Relapse: Durable clinical results are the prime objective in the successful treatment of tuberculosis and the achievement of this objective, or lack thereof, may be construed as a measure of the effectiveness of a given therapeutic procedure. The increment of relapses over a given period of time is one of the most important criteria of the effectiveness of therapy.

Roentgenological relapse of the disease was determined by the individual investigators in the 332 patients in this series (figure 10). As the percentage of relapse occurring in the four different regimens is cumulative and increases according to the length of time of observation from the beginning of therapy, it should be recalled that the four regimens were evaluated at different periods of time after the beginning of treatment. As patients on regimen D were observed for the longest period of time, the total increment of relapse should be the greatest in this group, unless this regimen was considerably more effective than any of the other three. The results in regimen D were evaluated approximately 100 days later than those in regimen A, and approximately one to two months later than those in regimens C and B. As the percentage of relapse is essentially the same for the four regimens, regimen D appears to be somewhat more effective in the prevention of relapse than the other regimens.

DISCUSSION

Although many problems concerning the most effective use of streptomycin in the treatment of pulmonary tuberculosis remain unsolved or only partially solved, a considerable amount of confirmatory and additional information has emerged since the American Trudeau Society streptomycin project was launched two years ago. Some of this information and better clarified concepts will be discussed.

The objective: The ideal and ultimate objective of an over-all therapeutic program is the securing of lasting arrest of the disease, preservation of adequate pulmonary function, prevention of permanent physical or mental deficiencies, and the restoration to the family and community of an individual who is as capable of carrying on in a competitive society as he was before the development of tuberculosis. The objective of streptomycin therapy must of necessity, however, be more circumscribed and limited. Under certain conditions streptomycin therapy may play only a minor role but in others a vital or decisive role in the over-all therapeutic program. In general and specific terms what should be the primary objective of streptomycin therapy? In general, it may

be considered that the primary objective of streptomycin is to bring into favorable balance vital resistance over susceptibility and dosage of organisms. Specifically, this is brought about by the inhibitory action of streptomycin on the tubercle bacillus.

Several questions immediately arise concerning the possible attainment of the general and specific objectives of streptomycin therapy which are especially important in relation to duration of treatment. (*1*) How rapidly does streptomycin exert a significant antimicrobial effect in clinical tuberculosis? (*2*) Are different degrees of antimicrobial effect obtained? (*3*) What are some of the more important factors that limit streptomycin effectiveness? (*4*) How long *should* effective antimicrobial therapy be maintained? (*5*) How long *can* effective antimicrobial therapy be maintained? (*6*) When and under what circumstances should other therapeutic procedures be used?

With regard to the first question, factors related to the drug, the infecting organism and the patient are all involved. The pharmacology of streptomycin has been thoroughly investigated and is well documented. It is known that the drug is rapidly absorbed into most of the body fluids but to what degree it is concentrated in different types of diseased and damaged tissue is not so well known. Just how rapidly the drug may exert a significant therapeutic effect upon pulmonary tuberculosis is dependent upon a number of factors, the most important of which are the type and extent of the disease. There is considerable evidence to show that therapeutic effect may be exerted quite rapidly under certain circumstances. For instance, rapid defervescence and disappearance of symptoms within forty-eight to seventy-two hours following the initial administration of streptomycin has been observed in patients acutely ill with high fever and other toxemic manifestations of recently developed tuberculous pneumonia. Rapid loss of symptoms referable to laryngeal, bronchial, or intestinal tuberculosis has also been observed within a very short time after the beginning of streptomycin treatment. Probably the most dramatic therapeutic effect of streptomycin is that sometimes noted in patients with tuberculous meningitis. McDermott *et al.* have reported upon the rapid therapeutic effect of streptomycin in both acute generalized lymphohematogenous tuberculosis and tuberculous pneumonia (8, 9).

If the drug has a more rapid and also greater therapeutic effect upon acute disease of recent origin, it could be argued that the duration of treatment for this type of pulmonary disease might be less than that for the subacute or more chronic forms of pulmonary tuberculosis. Moreover, it seems logical to assume that the relative stability of chronic disease is indicative of a more favorable balance between resistance and susceptibility and number of infecting bacilli. The corollary of this is that acute progressive disease is evidence of a relative lack of resistance or the presence of overwhelming dosage of organisms or of both. Rich has pointed out that there is no such thing as "solid immunity" or resistance adequate to prevent spread of the infection provided the dosage of organisms is sufficiently large (10). This fact is frequently demonstrated by the patient with chronic cavernous disease whose sputum is teeming with

tubercle bacilli yet who suffers no spread of disease for long periods of time until a brisk hemoptysis results in the aspiration of an overwhelming dose of bacilli into previously healthy areas of the lung with the development of tuberculous pneumonia. It may be concluded, therefore, that the presence of extensive, acute, predominantly exudative disease indicates a serious imbalance in the host-parasite relationship and is an important factor to be considered in the attainment of the objectives of streptomycin therapy.

Although resistance to tuberculosis can be measured imperfectly at best, it is known that certain patients may have greater actual and potential capabilities of coping with their disease than others. Even though the immediate and particularly the remote potentialities of a given lesion cannot be measured accurately, some idea of the ability of the patient to overcome the disease may usually be estimated. In the final analysis, these fundamental factors of dosage of organisms, patient resistance, the type, extent, and immediate and remote potentialities of the disease, and the possible effective use of other therapeutic procedures serve as guides and principles in the final determination of the proper duration of streptomycin treatment. Thus the objectives of streptomycin therapy may be realized within three or four weeks in some patients, in two to three months in others, and in still others they may never be realized.

Types of disease: The early clinical reports suggesting that recent acute exudative or lobular pneumonic lesions that have not progressed to extensive caseation respond exceedingly well to streptomycin have been confirmed by many investigators, notably those of the Veterans Administration, Army, Navy and the present group. At the January, 1948 meeting of the Chemotherapy Committee, Amberson and Stearns demonstrated illustrative cases of this type showing marked therapeutic effect of streptomycin. The comparative efficacy of streptomycin in different clinical types of disease is well shown in figure 2. Experience has also emphasized the fact that the earlier the drug is used for acute disease the more effective it is in the prevention of irreversible morphologic changes and consequent disturbances of function. It has also been the general experience that, despite rapid clearing of the acute inflammatory process, largely by resolution, many small presumably caseous and partially encapsulated foci remain. These residual foci may be the source of relapse or progression of the disease at some later date, particularly if adequate sanatorium or hospital treatment is not continued for a sufficient period of time.

The large majority of patients with fairly extensive lobular pneumonic disease usually have cavities which may or may not be roentgenologically demonstrable. Probably for this reason, "sputum conversion" without some other treatment in addition to streptomycin, even in patients with acute disease who show striking improvement, is usually not attained with any greater degree of consistency than in patients who have more subacute or chronic types of disease. This is well shown in figure 2.

Streptomycin has proved of great benefit in preparation of patients for thoracoplasty who have extensive exudative disease in addition to cavity formation. It is believed that in many such patients the disease would have continued to

progress and surgery would probably not have become feasible without streptomycin. Several illustrative cases of this type were shown by the group from New York State Tuberculosis Hospitals[7] at the January, 1948 Chemotherapy Committee meeting. Streptomycin has also been found to be of limited or considerable value in preparation of patients for surgery who have predominantly chronic fibrocaseous and cavernous disease with a subacute component and frequently a complicating tuberculous bronchitis. The partial, even though perhaps temporary, control of the more active pulmonary and bronchial disease is often sufficient to permit a relatively safe and effective surgical program. In addition, streptomycin may be of such marked benefit in patients with tuberculous laryngitis or enteritis, with consequent improvement in the general condition, as to allow successful thoracoplasty or other surgical procedures. On the other hand, streptomycin usually has little or no decisive therapeutic value when used alone and directed against chronic types of cavernous disease without recent or exudative components, or in the absence of tuberculous laryngitis, tracheobronchitis or enteritis.

Howlett and O'Connor have made detailed studies of the effect of streptomycin on chronic disseminated nodular tuberculosis of the lungs. At the meeting of the Chemotherapy Committee in January, 1948, they showed illustrative cases and pointed out that streptomycin may be strikingly effective in this type of chronic disease (11).

Methods and frequency of administration: Experience during the past two years has definitely shown that parenteral streptomycin therapy gives the best results. As a result of animal experimentation (6) and later clinical experience it was found unnecessary to administer the drug more often than twice daily, and once daily may be effective for certain patients. The maintenance of a high concentration of streptomycin in the blood is not necessary to achieve beneficial therapeutic results. A given daily dosage administered in four or five injections appears to cause more severe toxic reactions than the same amount administered once or twice daily. Streptomycin given by inhalation or aerosolization is much less effective, even for disease of the mucous membranes of the respiratory tract, than when administered parenterally. Streptomycin given by mouth is usually ineffective and is not recommended.

Daily dosage: Statistical data concerning the relative therapeutic effects of large versus small daily dosages have been presented above. These data would seem to indicate that the larger daily dosages resulted in more frequent as well as more marked improvement than the smaller daily dosages. It should be emphasized, however, that this finding appears to be somewhat at variance with the opinions and experiences of some of the individual investigators, including the writers, whose combined reports constitute the statistical data presented.

[7] Ray Brook State Tuberculosis Hospital, Ray Brook, New York, Dr. Harry A. Bray, Director; Homer Folks Tuberculosis Hospital, Oneonta, New York, Dr. Ralph Horton, Director; Hermann M. Biggs Memorial Hospital, Ithaca, New York, Dr. N. Stanley Lincoln, Director; Mt. Morris Tuberculosis Hospital, Mt. Morris, New York, Dr. Arthur M. Stokes, Director.

In an attempt to clarify this paradoxical finding the sources of probable clinical and statistical error have been sought and an endeavor made to evaluate their effects upon results obtained in the different regimens. Several factors, other than daily dosage, which may influence therapeutic results have already been discussed. Two important factors which significantly influence results in this series of patients are: (*1*) the unequal distribution of similar types of clinical material among the different regimens, and (*2*) the relatively small number of patients on all regimens, but especially on regimen B (46 patients).

Evaluation of the clinical material in each regimen shows that a much larger percentage of patients with predominantly *acute* disease are in regimens B and D than in A and C. The percentage distribution of acute disease in the four regimens is as follows: A, 36.3; B, 60.9; C, 40.4; and D, 52.1 (figure 1). It is generally agreed that the best results from streptomycin are usually attained in patients with recent acute, potentially reversible disease. This fact is strikingly shown in figure 2, in which it is seen that marked therapeutic improvement, measured in terms of clinical, roentgenographic, and over-all improvement, occurred approximately twice as often in patients with acute disease as in those with subacute disease. Moreover, improvement occurred from two and one-half to four times as often in patients with acute disease as in those with chronic disease. It is quite probable, therefore, that the superior results obtained in regimens B and D were more significantly related to the type of clinical material (predominantly acute disease) than to the difference in total daily dosage or the difference in duration of treatment. In this connection it should be recalled that results in regimen C were not as favorable as those in regimen B although patients on the former regimen were treated for the longer period of time. It is reasonable to assume, therefore, that the superior results on regimen B, as compared with those observed in the patients on regimen C, were most probably due to the larger percentage of patients with acute disease on regimen B, and to a less extent to the larger daily dosage given to the patients on this regimen. To recapitulate, it may be said that the most significant factors affecting results in this series of patients are: (*1*) the predominant type of clinical material treated with the individual regimens, (*2*) the total daily dosage, (*3*) the duration of treatment, and (*4*) the emergence of drug-resistant organisms, a phenomenon which is undoubtedly of the greatest significance in limiting the effectiveness of prolonged streptomycin treatment.

Duration of treatment: Before deciding upon the duration of streptomycin therapy for the individual patient, it is essential to consider the objectives, possibilities, and limitations of drug therapy. Effective antibacterial therapy is related to multiple factors, some primarily concerned with the patient, some with the disease, and others with the bacillus and the drug. It is also essential, in considering the duration of streptomycin treatment, to formulate an over-all therapeutic program that effectively integrates streptomycin therapy with other procedures.

If it were possible to eradicate all infection and there were no contraindications, it would be desirable under certain circumstances to continue streptomycin

therapy for whatever time required for the elimination of all viable organisms. As this is not possible and as there are a number of factors limiting the duration of effective therapy, the period of treatment selected for different types of pulmonary tuberculosis must be a compromise. The best compromise can be ascertained only after establishing a definite objective and careful weighing of all factors involved, immediate, remote and potential.

Emergence of streptomycin-resistant tubercle bacilli: Once the forces of vital resistance have become favorably balanced over dosage of organisms as a result of streptomycin therapy, the question arises as to how long should streptomycin be continued. If "drug-fast" organisms did not replace sensitive organisms in proportion to the duration of treatment and provided the drug produced no toxic effects, it probably would be wise to continue its use for an indefinite period of time on the principle that other therapeutic measures are used, namely, until the disease is well arrested. By adjusting the dosage of streptomycin on a basis of milligrams per kilogram of body weight, serious toxic effects usually can be avoided. It is not possible at present, however, to prevent the emergence of resistant strains of organisms, although there is some evidence to suggest that this occurrence may be delayed or reduced as a result of the simultaneous use of combined chemotherapy. It also appears that organisms become "drug-fast" regardless of the method or frequency of administration or the total daily dosage given. It has been well established that "drug-fastness" is definitely related to duration of treatment and that the incidence of resistant strains increases with prolongation of treatment. Organisms rarely become "drug-fast" before the end of the first month of streptomycin therapy. Usually, however, 75 to 80 per cent of patients discharge significantly "drug-fast" organisms by the end of the third or fourth month of treatment.

As streptomycin is usually essentially ineffective after the organisms become "drug-fast" and in view of the fact that pulmonary tuberculosis is characteristically a relapsing disease, the tremendous importance of avoiding the emergence of significant numbers of "drug-fast" organisms is obvious. If the primary objectives and limitations of streptomycin are kept in mind, and provided proper indications are observed and other therapeutic procedures are used at the opportune time, it is believed that the duration of streptomycin treatment may usually be sufficiently limited to avoid "drug-fast" organisms. There are certain exceptions to this statement. For example, it may be necessary to continue streptomycin for longer than four or six weeks for acute bilateral tuberculous pneumonia or for acute disseminated lymphohematogenous disease (nonmiliary) and under certain other circumstances in which there is no other effective therapy. Under such circumstances more prolonged treatment or retreatment seems justified, regardless of the emergence of some drug-fast organisms. The drug may continue to be of some limited value even though the majority of the infecting bacteria have become "fast." This reasoning is based upon the fact that scattered colonies of sensitive organisms are occasionally found in the presence of a large majority of resistant cells, or the reverse may be true.

It may be concluded, therefore, that optimum duration of treatment, just

as optimum dosage, is dependent upon weighing all factors involved, and in the final analysis must be decided upon in the light of the needs of the individual patient. This decision can best be made at the bedside.

Toxic effects: When given in relatively large daily dosage over prolonged periods, the toxic effects of streptomycin are second only in importance to the emergence of drug-fast organisms as a limiting factor in the usefulness of the drug in the treatment of tuberculosis. The early experiences (2, 7) showed that, when streptomycin was administered intramuscularly in 2.0 to 3.0 Gm. daily doses, it frequently caused disturbing and often severe toxic manifestations, the most serious being deafness and loss of labyrinthine function. Many other usually less significant reactions of a wide variety have also been observed.

As a result of the above findings, the present group explored many dosage schedules in an attempt to discover therapeutically effective but essentially nontoxic regimens. Daily dosage schedules ranging from 0.5 Gm. to 3.0 Gm. were tested. As mentioned above, few patients in the series received as much as 2.0 to 3.0 Gm. daily and the great majority received only 1.0 to 1.5 Gm. daily. With the smaller daily doses, the incidence and severity of toxic manifestations were significantly less. Deafness was not reported in any of the patients treated for pulmonary tuberculosis who are included in this report. Varying degrees of vestibular dysfunction, as evidenced by caloric stimulation tests and clinical symptoms or both, were observed fairly often. It should be mentioned that periodic caloric tests may show the presence of considerable depression of the labyrinthine function in the absence of symptoms. Depression of the labyrinthine function may disappear during treatment and it occasionally disappears after completion of treatment. Moreover, in patients having complete loss of labyrinthine function, the compensatory balancing mechanisms usually become sufficiently effective to prevent prolonged and marked disability. Although such patients may continue to improve for several months, difficulty in walking in the dark may persist indefinitely.

Serious toxic manifestations can be largely avoided or greatly minimized if careful attention is paid to the selection of the proper dosage schedule, which is best calculated on a milligram per kilogram body weight basis. In addition, periodic clinical and laboratory observations should be made, including caloric stimulation tests, audiograms and blood urea nitrogen determinations when indicated. Finally, toxic effects of any consequence are rarely encountered in patients receiving a daily dosage of 20 mg. per kg. of body weight. Although this dosage is undoubtedly beneficial in the majority of patients when proper indications are observed, nevertheless there is considerable evidence to suggest that a daily dosage of 30 to 40 mg. per kg. of body weight perhaps exerts a greater therapeutic effect, at least in patients with certain types of disease. As dihydrostreptomycin appears to be of lower toxicity than streptomycin in daily dosages of 30 to 40 mg. per kg. of body weight, it is believed that it may be possible to provide greater antimicrobial effect with the derivative than with the parent drug (12). In any event, it has been conclusively demonstrated that therapeutically effective and essentially nontoxic dosage schedules of streptomy-

cin can be determined with considerable accuracy for the types of pulmonary tuberculosis which usually respond favorably to the drug.

Relapse: There is no theoretical reason to assume, or experimental, pathological or clinical evidence to indicate, that streptomycin, especially when used without collapse therapy, should or will reduce the incidence of relapse. On the contrary, it is logical to believe that the opposite may prove to be true. If the term relapse is used in a broad sense to denote progression of disease that has become clinically or roentgenologically stabilized or regressive as a result of streptomycin therapy, or the reactivation of disease that has become arrested, there is considerable reason to believe that relapse is more likely to occur in patients who have received streptomycin. The reasons for this belief are: (*1*) Many patients who do not receive streptomycin will not improve and therefore will not "qualify" or become candidates for relapse for their disease may steadily progress until death ensues. (*2*) Conversely, many patients who receive streptomycin will improve without attaining closure of cavities or reversal of infectiousness. In such instances the disease will usually relapse, particularly if collapse therapy or some other definitive type of treatment is not used at the opportune time. (*3*) Patients who improve without streptomycin do so because of the re-establishment of a favorable host-parasite relationship as a result of an autogenous mechanism. Patients who improve as a result of autogenous mechanisms are more likely to attain lasting results than are patients who have inferior resistance and improve as a result of streptomycin therapy, an exogenous mechanism. (*4*) There is no evidence to indicate that streptomycin, even when it produces striking improvement, increases the native resistance of the patient, an important factor in the incidence of relapse.

For these and other reasons previously mentioned which clearly indicate that streptomycin is usually not a definitive form of therapy, additional treatment at the opportune time is essential if relapse is to be minimized or avoided. Relapse may occur (*a*) during treatment, (*b*) soon after completion of treatment, or (*c*) with the emergence of resistant organisms. Therefore, it is evident that, if collapse therapy or other surgery is to be used most effectively and under the most favorable circumstances, it must be instituted in many instances within a few weeks after streptomycin is begun. Several of the present group of investigators have reported that many patients with extensive and recently acute, predominantly exudative disease have been able to have extensive surgery with remarkably little difficulty and with excellent clinical and bacteriological results.

SUMMARY

The results of streptomycin therapy in 332 patients with pulmonary tuberculosis, treated by a group of cooperating investigators in the American Trudeau Society, are reported in summary form.

On the basis of clinical, roentgenologic, and bacteriologic observations, it was clearly demonstrated, as previously reported by others, that the degree of therapeutic efficacy of streptomycin is definitely related to the *character* of the pulmonary disease. Although improvement occurred much more frequently

and was more marked in patients with predominantly acute disease than in those with predominantly chronic disease, nevertheless significant improvement was reported by most of the investigators in a small percentage of patients with chronic disease. As the progression of *chronic* tuberculosis is fundamentally related to the occurrence of *acute* episodes, the great potentialities of the strategic use of short courses of streptomycin to combat such episodes should be stressed.

The larger daily dosages of streptomycin appeared to bring about more frequent and more marked improvement than the smaller daily dosages. Even though the larger daily dosages appeared to produce greater therapeutic effect, this advantage was often minimized or nullified by the more severe toxic manifestations incident to larger dosage. The availability of dihydrostreptomycin, which appears to be less neurotoxic than streptomycin, may aid in the solution of this problem.

The occurrence of drug-fast organisms was found to be definitely related to duration of treatment. Organisms resistant to 10 or more γ of streptomycin rarely occurred in patients treated for one month, but appeared frequently in patients treated for two months or more. It appears that "drug-fastness" is unquestionably a significant factor in limiting the effectiveness of the antibiotic.

The failure to close cavities and convert sputum in a larger percentage of patients and the relapse of the disease with increasing frequency with passage of time (patients observed six to ten months after the beginning of treatment) suggest the limitations of antibacterial therapy for this characteristically chronic disease and emphasize the absolute necessity of also using more definitive treatment, notably collapse therapy, if lasting results are to be achieved.

SUMARIO

Obra de Investigación de la Estreptomicina en la Tuberculosis por la American Trudeau Society. Memoria Sumarizada

Esta memoria sumariza los resultados de la estreptomicinoterapia en 332 tuberculosos pulmonares tratados en cooperación por un grupo de investigadores de la American Trudeau Society.

A base de las observaciones clínicas, radiológicas y bacteriológicas, demostróse claramente, según ya han comunicado otros autores, que la eficacia terapéutica de la estreptomicina guarda relación bien definida con la *naturaleza* de la afección pulmonar. Aunque la mejoría fué mucho más frecuente y más pronunciada en los casos predominantemente agudos que en los predominantemente crónicos, la mayor parte de los investigadores comunicaron tembién mejoria significativa en un pequeño porcentaje de los enfermos crónicos. Como la agravación de la tuberculosis *crónica* se relaciona fundamentalmente con la ocurrencia de episodios *agudos*, hay que recalcar las grandes potencialidades que entraña el empleo estratégico de breves series de estreptomicina para combatir dichos episodios.

Las dosis diarias mayores de estreptomicina obtuvieron al parecer mejorías más frecuentes y más notables que las dosis diarias más pequeñas. Si bien las primeras produjeron aparentemente mayor efecto terapéutico, esta ventaja

fué a menudo atenuada o borrada por las manifestaciones tóxicas mas graves que las acompañaron. En la solución de este problema tal vez ayude la introducción de la dihidroestreptomicina, que parece ser menos tóxica que la estreptomicina.

La aparición de microbios estreptomicinorresistentes resultó hallarse netamente relacionada con la duración del tratamiento. Microbios resistentes a 10 o más γ de estreptomicina fueron raramente observados en los enfermos tratados por un mes, pero aparecieron frecuentemente en los tratados por dos meses o más. Según parece, la farmacorresistencia constituye indudablemente un factor importante en la limitación de la efectividad del antibiótico.

El fracaso en lo relativo a clausura de las cavernas y viraje del esputo en un porcentaje mayor de enfermos y las recidivas cada vez más frecuentes con el transcurso del tiempo (en los sujetos observados de seis a diez meses desde el comienzo del tratamiento) indican las limitaciones de la antibacterioterapia en esta enfermedad tan típicamente crónica y recalcan la necesidad abosulta de usar además un tratamiento más definitivo, notablemente la colapsoterapia, si se van a lograr resultados duraderos.

Acknowledgments

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CONSTITUTIONAL FACTORS IN RESISTANCE TO INFECTION[1, 2]

I. The Effect of Estrogen and Chorionic Gonadotropin on the Course of Tuberculosis in Highly Inbred Rabbits

MAX B. LURIE, SAMUEL ABRAMSON[3] AND MARVIN J. ALLISON

Introduction

In a study (1) on the native resistance of inbred rabbit families of varying hereditary resistance to tuberculosis it was found that the fundamental variant in the disease developed by these families was the degree of localization of the infection at the portal of entry. Families of high resistance effectively limited the process to the lung, if the tuberculosis was of natural respiratory origin, with little or no dissemination of the infection by hematogenous or lymphogenous routes. In families of low resistance, on the other hand, the primary tuberculosis in the lung progressed rapidly and was soon widely disseminated through the body by the vascular system. As bacterial infection takes place chiefly in the connective tissue, it was thought that the permeability of this tissue to particulate matter might be one of the factors in this resistance. In fact, in some families there is a parallelism between the spread of tuberculosis and the spread of India ink in the connective tissue of the skin. That the spread of particulate matter in the skin might be under the influence of naturally occurring hormones was suggested by the observation that male rabbits (2) limited this spread more effectively than females. Furthermore, Sprunt (3) has shown that the administration of estrogen restricted the spread of India ink in the skin. On the other hand, the intravenous injection of chorionic gonadotropin (4), which induced the formation of corpora lutea in the ovaries and the secretion of progesterone, enhanced this spread.

It would follow, therefore, if the spread of particles, and among them tubercle bacilli, is a factor in the native resistance to tuberculosis, that suitable treatment of animals with estrogen should retard the tuberculous process. Conversely, appropriate exposure of rabbits to chorionic gonadotropin or progesterone should enhance the disease.

In this and the subsequent studies an endeavor was made to test this assumption. In general, the observations made substantiate the expected results. Analysis of the results, however, has led beyond the point of departure of the effect of these hormones on the spread of particles to the problem of their influence on the tuberculin sensitivity of the skin and on the allergy of the internal organs; to their action on the irritability of the skin to noxious agents in general; and to their role in the occurrence of amyloid degeneration. The effect of these hormones on the circulating lymphocytes, on the adrenal, and on antibody

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[2] Aided by a grant from the Commonwealth Fund.

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production was studied with the view of a possible interrelation between these hormones and the functions of the pituitary gland and the adrenal cortex.

Materials and Methods

Female litter mate pairs of the susceptible family C (1) of the same age and of identical genetic and environmental history for the past six to eight inbred generations were divided into two groups. One-half served as controls and received 0.5 cc. of sterile sesame oil subcutaneously once a week. The other litter mates were given 0.5 mg. α of estradiol dipropionate[4] in 0.5 cc. sesame oil at the same time and by the same route. Rabbit pairs older than 10 months were ovariectomized, divided into control and experimental groups, and treated in the same manner. After the experimental rabbits had been under the influence of the estrogen for about two weeks, 0.5 cc. of 1:5 dilution of autoclaved India ink in saline was injected intracutaneously into both groups. The spread of the particulate matter in the skin was determined on the next day by planimeter in a manner previously described (4). The results will be discussed in the last paper of this series. When the estrogenic effect in the experimental animals was indicated by a marked increase in the size and congestion of the vulva and a definite enlargement of the nipples, these rabbits, together with their litter mate controls, were infected intracutaneously with the same dose of virulent bovine tubercle bacilli of the Ravenel strain. The inoculations were made on one side of the chest wall at the same distance from the midline and from the axilla. As the number of suitable litter mate pairs from these inbred families available at any one time was small, it was necessary to limit each individual experiment to 4 paris and to repeat the experiment three times. The infecting dose varied in each group and was determined by colony counts of the inoculum (5). Needless to say, in each experiment the control and estrogen-treated rabbits received the same dose. The estrogen and sesame oil injections, respectively, were continued at weekly intervals throughout the course of the disease.

Two additional similar experiments were performed with litter mate pairs of the ninth and tenth inbred generations of the more resistant family A. The special conditions obtaining in these will be detailed later.

Two and seven days after inoculation, and at weekly intervals thereafter until the death of the animal, the skin lesion at the site of infection was measured and its volume determined according to a formula previously described (1). Likewise, the size of the axillary lymph nodes draining the site of inoculation was followed throughout the course of the disease.

Whenever one of a pair died, its litter mate was killed and the extent of the disease in the draining lymph nodes and lungs was determined objectively by their weight in grams. A careful search of all the organs was made for disseminated lesions and their character and magnitude was estimated. The weight of the uterus and vagina, the adrenals, the pituitary and other organs was determined.

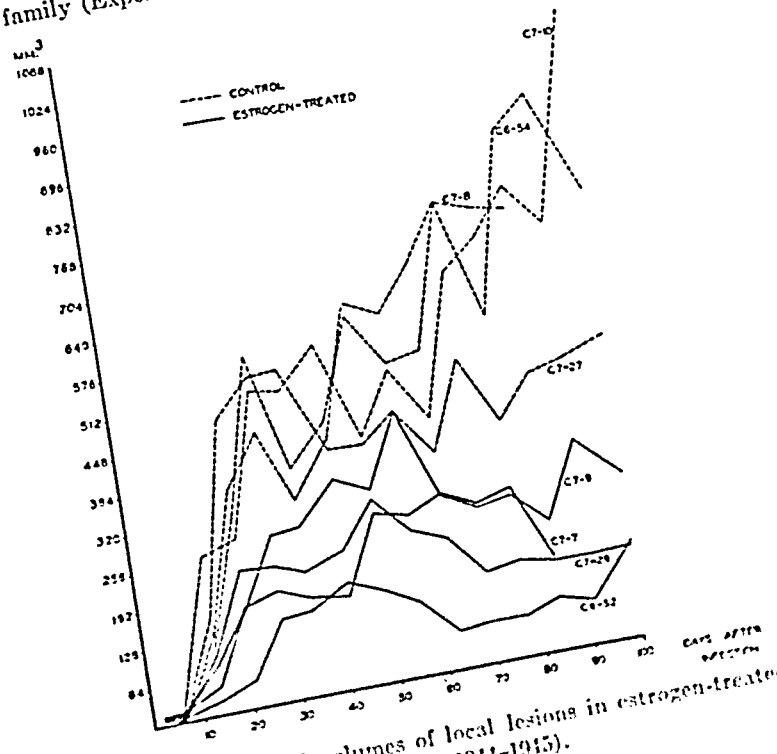
Observations

The effect of estrogen on tuberculosis

In text figure 1 may be seen the average volume of the lesion at the site of inoculation in the skin throughout the course of the disease in estrogen-treated

[4] This and other sex hormones were generously furnished by Dr. Ernst Oppenheimer of the Ciba Pharmaceutical Products, Inc.

TEXT FIGURE 1. Average volume of local lesion in estrogen-treated and control litter mates of the C family (Experiment of 1944–1945).



TEXT FIGURE 2. Individual volumes of local lesions in estrogen-treated and control litter mates of the C family (Experiment of 1944–1945).

and control litter mates of the first experiment on the C family. The infecting dose was 82,000 viable bacilli. In text figure 2 is plotted the individual volumes of these lesions in each of these 8 rabbits during the entire course of the infection. It is evident that the progress of the lesion at the site of inoculation is markedly and uniformly retarded in the estrogen-treated rabbits as compared with that in their litter mate controls.

TABLE 1

The effect of estrogen on the progress of tuberculosis in litter mate pairs of the inbred, susceptible family C; experiment 1944–1945

CONTROL, C OR EXPERIMENTAL, E	RABBIT NUMBER	PARENTS AND AGE	DIED D, KILLED K	VOLUME OF LESION AT SITE OF INOCULATION ON 28TH DAY AFTER INFECTION	WEIGHT OF UTERUS AND VAGINA	WEIGHT OF TUBERCULOUS NODES DRAINING SITE OF INFECTION	EXTENT OF DISEASE IN LUNGS AND THEIR WEIGHT	EXTENT OF DISEASE IN PLEURA	EXTENT OF DISEASE IN KIDNEYS	NUMBER OF OTHER ORGANS METASTASIZED
		months		*mm.³***		*grams*	*grams*			
C	C6-54*	C5-52 x C5-40 10.6	D	578	6.0	1.8	+++ 39.7	+++	++	2
E	C6-52*	C5-52 x C5-40 10.6	K	140	36.6	2.1	+ 12.3	±	±	1
C	C7=27	C5-52 x C6-40 4.0	D	544	9.3	4.7	++++ 52.1	++±	+++	8
E	C7=26	C5-52 x C6-40 4.0	K	228	16.7	2.5	++ 19.1	+±	++	4
C	C7-8	C6-11 x C6-40 4.0	K	452	7.2	4.7	++++ 47.3	++	+++	6
E	C7-7	C6-11 x C6-40 4.0	D	187	13.6	1.2	+++ 43.5	++	++	2
C	C7-10	C6-11 x C6-40 4.0	K	520	5.8	4.7	+++ 38.8	++++	+++	10
E	C7-9	C6-11 x C6-40 4.0	K	280	19.8	2.3	+++ 33.2	+++	+++	6

* Ovariectomized on sixth and seventh day following infection.
** Per cubic centimeter of free flowing venous blood.

In table 1 are presented the essential data at the time of death, which, it will be remembered, was the same for the two rabbits of each pair. In column 3 the parents and age of each of these pairs are recorded. They are identical in each instance. The volume of the lesion at the site of inoculation on the twenty-eighth day of infection, listed in column 5, again shows how uniformly the growth of the lesion in the estrogen-treated rabbits was retarded. That the estrogen exercised its physiological effect on the experimental animals is shown in column 6, where the weight of the uterus and vagina of each of the estrogen-treated rabbits is seen to be approximately two to six times that of its litter mate con-

trol. The extent of the disease in the lymph nodes draining the site of inoculation and the tuberculosis in the lungs in general is less in the experimental than in the control animals. The same is true for the remaining organs. In each case, however, the degree of retardation varies; in some it is marked, in others it is less pronounced. The most consistent observation in this and in subsequent experiments of the same character appears in the last column where it may be seen that the number of metastases to other organs is uniformly less in the estrogen-treated than in their control litter mates. It may be noted that ovariectomy did not diminish the retarding effect of estrogen on the progress of the disease.

In text figure 3 may be seen the average progress of the lesion at the site of inoculation in 4 estrogen-treated rabbits and in 4 litter mate controls of the second experiment on the C family, where the infecting dose contained 162,000 living microorganisms. Again it is clear that estrogen markedly retarded the growth of the lesion at the portal of entry throughout the course of infection. Figures 1 and 2 (Plate I) illustrate this effect in the two ovariectomized litter mates, C7-13 and C7-12, during the fourteenth week of the infection. The larger size of the nipple of the latter attests to the estrogenic influence to which it was subjected.

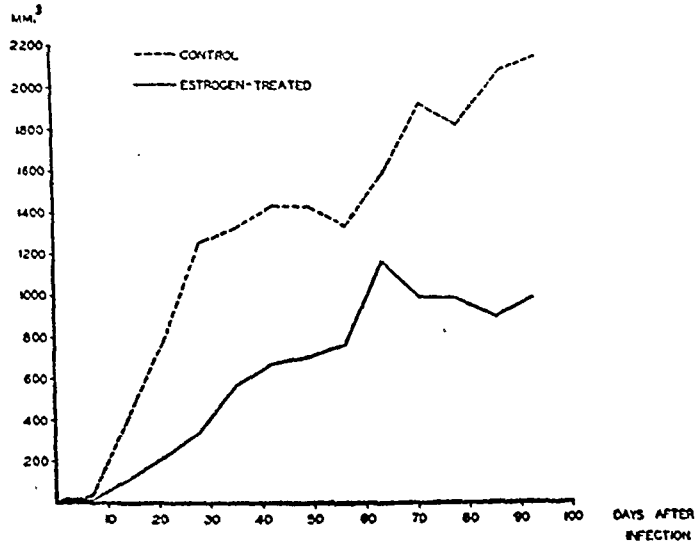
Table 2 presents in detail the extent of tuberculosis in the lung and in the other organs at the time of death, as well as other pertinent data listed in the previous experiment. It is noteworthy that all of the control rabbits died, whereas all the estrogen-treated mates were killed. Figures 5, 6, 7 and 8 (Plate II) depicit the tuberculosis in the lung and kidney and the development of the uterus and vagina in each of these 4 pairs at autopsy. It is clear that in every instance where the rabbit was under the influence of estrogen, as indicated by the larger size of the uterus, the disease was less extensive than in its litter mate control. Again the retarding effect of estrogen was pronounced in some and less marked in others. As in the last experiment, the number of organs metastasized was conspicuously reduced in the estrogent-reated animals.

In an effort to determine whether estrogen treatment will effectively suppress an infection with small numbers of virulent tubercle bacilli, a third group of 4 litter mate pairs of the susceptible family C was inoculated intracutaneously with about 10 virulent tubercle bacilli, as determined by culture. The progress of the lesion at the site of inoculation in the control and estrogen-treated rabbits is presented in text figure 4.

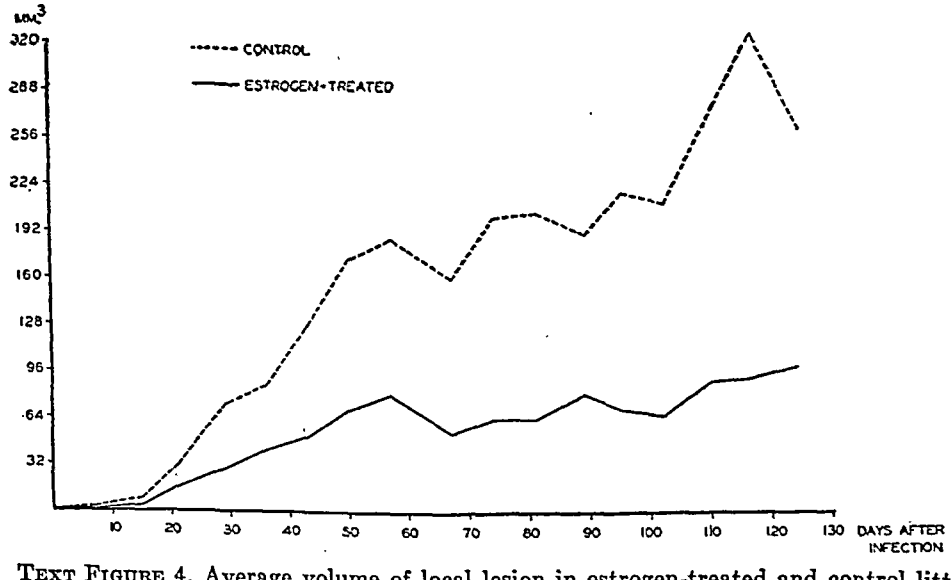
The striking effect of bacillary dosage on the rate of growth of the cutaneous lesion both in the control and estrogen-treated rabbits is evident by comparing the progress of the lesions in this group with that in the two previous experiments as recorded in text figures 1 and 3, respectively. The delay in the development of the lesion with the smaller dose is pronounced. Nevertheless despite this very slow development of the disease, estrogen treatment only retarded its progress, but did not suppress it.

The results are presented in detail in table 3. It may be noted that in 3 of the 4 pairs estrogen definitely slowed the progress of the disease in the internal

organs and reduced its dissemination. In one of these pairs, however, the tuberculosis in the estrogen-treated litter mate was actually more extensive than in the control. It is noteworthy that this pair was quite young and had not



TEXT FIGURE 3. Average volume of local lesion in estrogen-treated and control litter mates of the C family (Experiment of 1945–1946).



TEXT FIGURE 4. Average volume of local lesion in estrogen-treated and control litter mates of the C family (Experiment of 1946–1947).

attained sexual maturity at the beginning of the experiment. While the uterus and vagina of all the estrogen-treated rabbits were quite hypertrophied, these organs were also quite large in some of the control litter mates. This is possibly

TABLE 2

The effect of estrogen on the progress of tuberculosis in litter mate pairs of the inbred, susceptible family C; experiment 1945-1946

CONTROL, C OR EXPERIMENTAL, E	RABBIT NUMBER	PARENTS AND AGE	DIED D, KILLED K	VOLUME OF LESION AT SITE OF INOCULATION ON 28TH DAY AFTER INFECTION	WEIGHT OF UTERUS AND VAGINA	WEIGHT OF TUBERCULOUS NODES DRAINING SITE OF INFECTION	EXTENT OF DISEASE IN LUNGS AND THEIR WEIGT	EXTENT OF DISEASE IN PLEURA	EXTENT OF DISEASE IN KIDNEYS	NUMBER OF OTHER ORGANS METASTASIZED
		months		*mm.³***		*grams*	*grams*			
C	C7-36	C6-11 x C6-40 4.8	D	1,360	6.3	2.5	+++++ 67.6	++	++++	16
E	C7-40	C6-11 x C6-40 4.8	K	408	22.2	3.75	++± 32.5	+++	+++	8
C	C7-16*	C6-11 x C6-40 12.9	D	926	7.9	7.3	+++++ 74.4	++	++±	4
E	C7-15†	C6-11 x C6-40 12.9	K	486	20.7	1.95	+++ 43.5	+	+	3
C	C7-29	C6-11 x C6-40 7.3	D	1,544	7.2	0.96	++± 27.2	++±	++	8
E	C7-28	C6-11 x C6-40 7.3	K	345	25.7	1.07	± 10.3	+++	+	
C	C7-13*	C6-11 x C6-40 12.9	D	1,207	6.5	3.55	+++++± 56.5	+++++	+++	7
E	C7-12*	C6-11 x C6-40 12.9	K	240	27.8	3.35	++ 20.0	+	+±	4

* Ovariectomy performed three weeks before inoculation.

† Rabbit had long standing infection of subcutaneous tissues which persisted for 70 days during the course of the infection.

** Per cubic centimeter of free flowing venous blood.

accounted for by the fact that these females were inadvertently exposed to males fourteen to nineteen days prior to the determination of their uterine weights. A fourth trial with estrogen was made on 4 litter mate pairs of the tenth in-

PLATE I

FIG. 1. (Upper left). Lesion at site of inoculation in skin of control rabbit C7-13 on fourteenth week of infection. The large ulcerated lesion is seen in upper half of photograph with extension toward the abdomen below it. The large draining axillary node is seen in the left upper portion of the figure. The nipple is below the label and to the left.

FIG. 2 (Upper right). Lesion at site of inoculation in skin of estrogen-treated rabbit C7-12 (litter mate of C7-13, shown in figure 1), on the fourteenth week of infection. A small lesion with little spread is seen in the lower half of the figure. Compare the size of the nipple in this rabbit, seen above the ulcer, with that of its litter mate in figure 1.

FIG. 3 (Lower left). Lesion at site of inoculation in skin of control rabbit A9 = 51 on the 128th day of infection. The well-defined, raised lesion with its small ulcer is seen in the upper left portion of the photograph.

FIG. 4 (Lower right). Lesion at site of inoculation in skin of gonadotropin-treated rabbit A9 = 56 (litter mate of A9 = 51, shown in figure 3) on the 128th day of infection. The large ulcer, with numerous metastases in the skin, and the extensive involvement of the draining lymph nodes are readily seen.

PLATE I

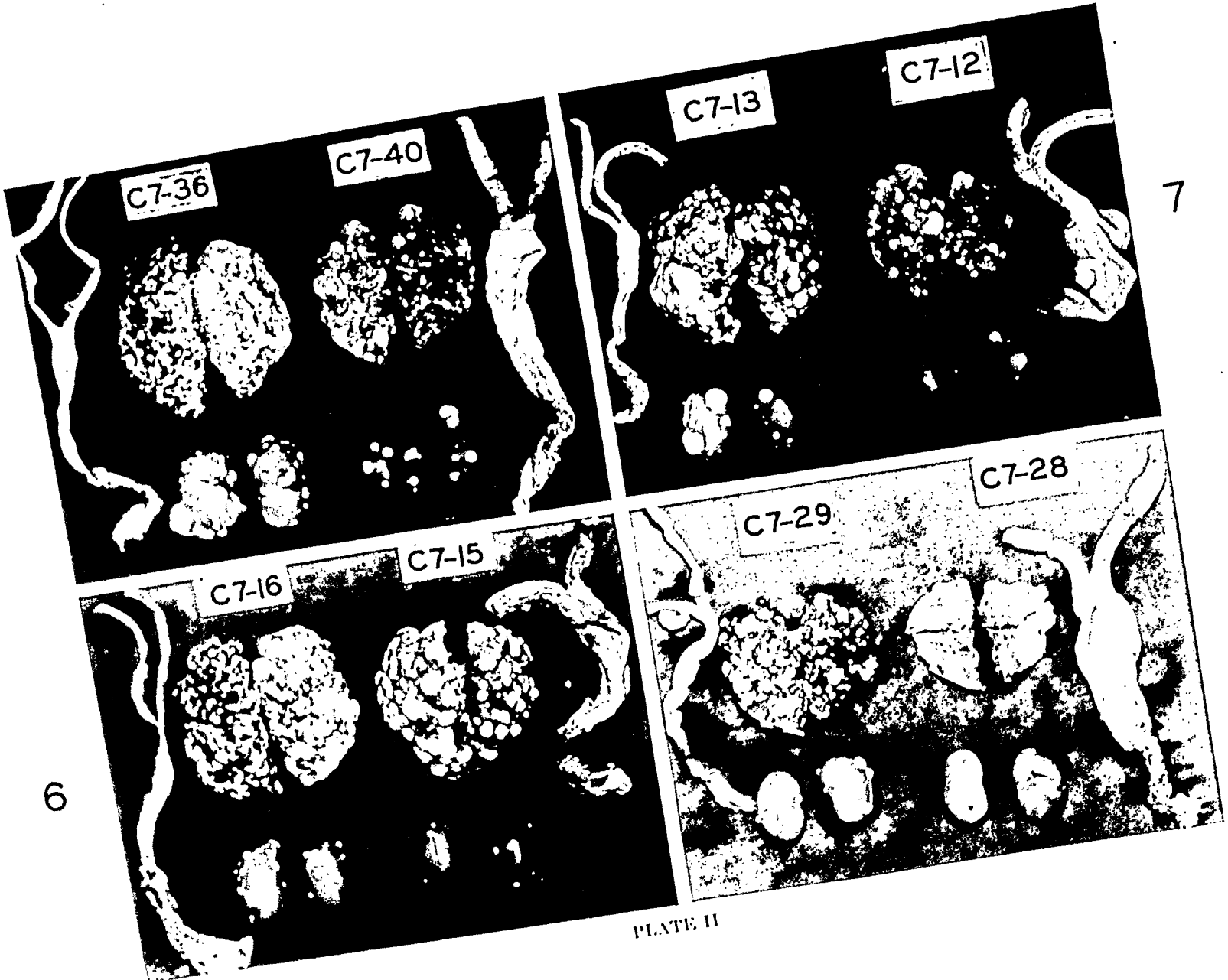


PLATE II

TABLE 2

The effect of [illegible] on the progress of [illegible] in the mean [illegible] of the [illegible] [illegible] (Experiment [illegible])

[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]

the retarding effect of estrogen on the growth of the lesion at the site of inoculation is again confirmed. In all but one of the pairs the extent of the disease in the lungs and the number of metastatic foci were less in the estrogen-treated

TABLE 4

The effect of estrogen on the progress of tuberculosis in litter mate pairs of inbred family A; Experiment 1946–1947

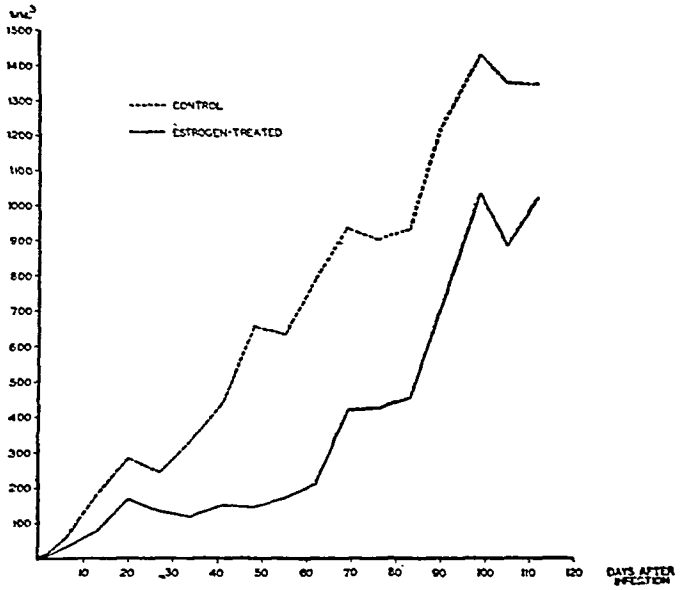
CONTROL, C OR EXPERIMENTAL, E	RABBIT NUMBER AND SEX, PARENTS, AGE IN MONTHS AND WEIGHT IN GRAMS AT BEGINNING OF EXPERIMENT	DIED D, KILLED K	VOLUME OF LESION AT SITE OF INOCULATION ON 41ST DAY AFTER INFECTION	WEIGHT OF UTERUS AND VAGINA	WEIGHT OF TUBERCULOUS NODES DRAINING SITE OF INFECTION	EXTENT OF DISEASE IN LUNGS AND THEIR WEIGHT	EXTENT OF DISEASE IN PLEURA	EXTENT OF DISEASE IN KIDNEYS	NUMBER OF ORGANS METASTASIZED BEYOND SITE OF INOCULATION
			cm.³**		grams	grams			
C	A10=19 ♂ A9=86 x A8=99 6 months; wt. 2,750 Gm.	D	823	—	5.4	++++ 36.6	++++	+++	15
E	A10=16 ♂ A9=85 x A8=99 6 months; 2,850 Gm.	K	112	—	3.3	++ 16.2	++	+	5
C	A10=23 ♂ A8=85 x A8=99 3.3 months; 2,250 Gm.	K	202	—	14.3	++ 17.0	+	+	8
E	A10=21 ♂ A9=86 x A8=99 3.3 months; 1,850 Gm.	D	150	—	—	+++ 43.7	+++	++	11
C	A10=26 ♀ A9=125 x A8=99 4 months; 2,650 Gm.	D	320	7.8	—	+++ 31.7	0	+	13
E	A10=25 ♀ A9=125 x A8=99 4 months; 2,400 Gm.	K	151	12.2	8.5	+++ 27.3	++	++	11
C	A10=30 ♀ A9=125 x A8=99 4 months; 2,450 Gm.	D	420	7.7	3.0	++++ 69.1	++	++++	11
E	A10=27 ♀ A9=125 x A8=99 4 months; 2,250 Gm.	K	179	30.7	2.35	++++ 57.6	+	++	7

** Per cubic centimeter of free flowing venous blood.

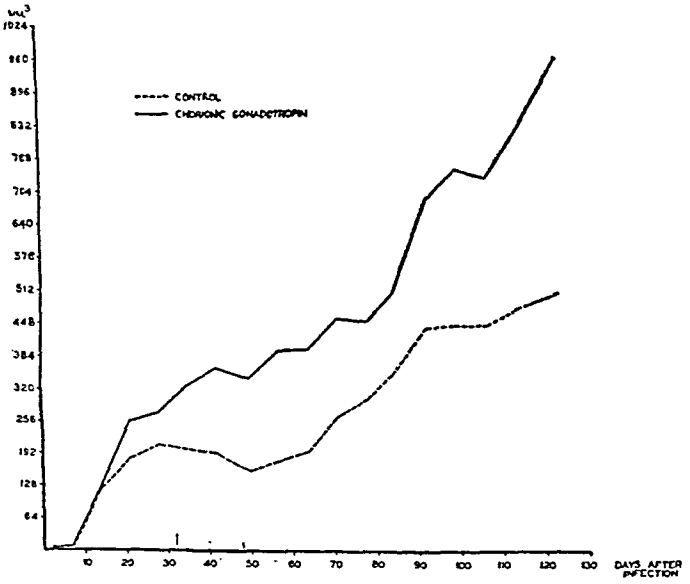
animals. The single litter mate in which the estrogen failed was only 3.3 months old at the beginning of the experiment.

The results of the four experiments are summarized in table 5. The pairs are listed in order of the age of the rabbits at the beginning of the experiment. The extent of the disease in the lung, pleura and kidney is given a single numerical

value, which is the sum of the degree of "plus" involvement of each, one plus being assigned the value of one.



TEXT FIGURE 5. Average volume of local lesion in estrogen-treated and control litter mates of the A family (Experiment of 1946).



TEXT FIGURE 6. Average volume of local lesion in chorionic gonadotropin-treated and control litter mates of the A family (Experiment of 1943).

It is clear that estrogen retarded the progress of the disease at the site of inoculation in the skin of all rabbits. In 13 out of the 16 pairs tested the estrogen definitely reduced the extent of the disease in the lung, pleura and kidney, which

are the usual seats of the most extensive lesions, and diminished the number of foci disseminated by lymphogenous and hematogenous routes. Ovariectomy did not influence this beneficial effect of estrogen. In one pair aged 4 months, while the number of metastases was reduced in the estrogen-treated mate, the extent of the disease in the above organs was greater. In two pairs aged 3.3 and 4.2 months, respectively, estrogen appeared to exert an enhancing effect on the extent and dissemination of the disease. Furthermore, in 12 of these pairs, the experiment was continued until the death of one of the litter mates. The remaining 4 pairs were killed. It is noteworthy that in the first group 10 of the control rabbits succumbed before the estrogen-treated mate. Only two experi-

TABLE 5

The effect of estrogen on the progress of tuberculosis in litter mate pairs according to their age at the beginning of the experiment

CONTROL					ESTROGEN-TREATED				
Litter mate pair	Age at beginning of experiment	Extent of disease in lung, pleura and kidney	Number of metastic foci	Killed K, died D	Litter mate pair	Age at beginning of experiment	Extent of disease in lung, pleura and kidney	Number of metastatic foci	Killed K, died D
	months					*months*			
(1)	12.9*	9.5	4	D	(1)	12.9*	5.0	3	K
(2)	12.9*	12.5	7	D	(2)	12.9*	4.5	4	K
(3)	12.8*	12.0	13	D	(3)	12.8*	7.0	11	K
(4)	10.6*	8.0	2	D	(4)	10.6*	2.0	1	K
(5)	7.3	7.0	8	D	(5)	7.3	4.5	2	K
(6)	6.0	9.5	15	D	(6)	6.0	4.0	5	K
(7)	5.8	7.0	12	K	(7)	5.8	4.5	7	K
(8)	5.8	4.5	6	K	(8)	5.8	3	4	K
(9)	4.8	11.0	16	D	(9)	4.8	8.5	8	K
(10)	4.2	1.5	4	K	(10)	4.2	5.5	6	K
(11)	4.0	11.0	11	D	(11)	4.0	7.0	7	K
(12)	4.0	9.5	8	D	(12)	4.0	5.5	4	K
(13)	4.0	9.0	6	K	(13)	4.0	7.0	2	D
(14)	4.0	10.0	10	K	(14)	4.0	9.0	6	K
(15)	4.0	3.5	13	D	(15)	4.0	6.5	11	K
(16)	3.3	2.5	8	K	(16)	3.3	8.5	11	D

* Ovariectomized.

mental animals died before their controls and these were 3.3 and 4 months of age, respectively, at the beginning of the experiment. It is plain, therefore, that estrogen uniformly retarded the tuberculosis of animals beyond the age of sexual maturity. Before that age the effect of estrogen was not as constant. The possible significance of this observation will be discussed in the last paper of this series.

THE EFFECT OF CHORIONIC GONADOTROPIN, PROGESTERONE AND OVARIECTOMY ON TUBERCULOSIS

Since chorionic gonadotropin increases the spread of particles in the skin (4) and thus exercises an effect opposite to that of estrogen, it is obvious, if spread

is a factor in resistance to tuberculosis, that the appropriate administration of this hormone should enhance the tuberculous process.

Materials and Methods

Accordingly, the following experiment on female rabbits was set up. Litter mates, or rabbits descended from at least one common parent of the seventh to the ninth inbred generations of the resistant A family, were divided into two groups. One group served as control; the others were given 0.02 to 0.2 mg. chorionic gonadotropin (6) intravenously. About one week later, when about 10 actively functioning corpora lutea had developed in the ovaries of similarly treated rabbits, both groups were injected intracutaneously with the same suspension of virulent bovine tubercle bacilli of the Ravenel strain. The control animals received no treatment. The experimental animals continued to receive the gonadotropin intravenously every tenth day throughout the course of the disease, in order to induce fresh crops of corpora lutea at a time when the preceding secretory bodies were beginning to undergo regression.[5] The progress of the lesion at the site of inoculation was carefully measured throughout the course of the infection. Some pairs were allowed to die from their disease in order to determine the duration of the disease in the two groups. In other instances, if one of a pair died its mate was killed on the same day. The extent of the disease in the various organs was carefully ascertained both macroscopically and microscopically.

In text figure 6 may be seen the average progress of the lesion at the site of inoculation throughout the course of the disease in 4 rabbits under the influence of gonadotropin as compared with that in 4 untreated litter mates. It is clear that the volume of the lesion in the rabbits under the influence of the gonadotropin is consistently larger than that of the control rabbits.

In table 6 are listed all the pertinent data of the above 4 pairs as well as those of additional control and gonadotropin-treated animals studied on another occasion. For the sake of brevity they are placed together in one table. All the rabbits were sexually mature and thus were capable of fully responding to the gonadotropin. The close genetic relationship, if not identity, of these two groups is shown in column 3. That the progress of the lesion at the site of inoculation was uniformly enhanced in the gonadotropin-treated animals is shown in column 4 and illustrated in figures 3 and 4 (Plate I). In the majority of instances the extent of the disease in the internal organs was definitely enhanced and in 6 of the 7 pairs of experimental animals the degree of dissemination of the disease by hematogenous and lymphogenous routes was definitely greater than in the controls. It is noteworthy that there was a definite tendency for a shorter duration of the disease in the experimental animals. That the gonadotropin-treated animals were under the physiologic influence of the hormone is shown in the last column of this table, where it may be seen that all the gonadotropin animals showed either corpora lutea in the ovary or diffuse luteinization of its stroma.

That these results are specific for gonadotropin and that the observed differ-

[5] We are indebted to Dr. Gregory Pincus who suggested this procedure and to Dr. Samuel Gurin who furnished the highly purified gonadotropin.

ences could not be attributed to the varying natural resistance of these rabbits was shown by the following observations. Four litter mates of the first 4 pairs listed in table 6 were given daily subcutaneous injections of 1 to 2 mg. of progesterone together with 5 to 10 γ of estradiol (7) for two weeks before infection and every day thereafter throughout the course of the disease. It was found that this treatment exerted no discernible effect either on the progression of the lesion at the site of inoculation, the extent of the disease in the internal organs, or the dissemination of the tuberculosis by vascular channels as compared with

TABLE 6

The effect of chorionic gonadtropin on the course of tuberculosis in female rabbits of the highly inbred A family

GROUP	RABBIT NUMBER AND AGE IN MONTHS	PARENTS	VOLUME OF LOCAL LESION SIX WEEKS AFTER INFECTION	EXTENT OF PULMONARY LESIONS	EXTENT OF PLEURAL LESIONS	EXTENT OF RENAL LESIONS	SURVIVAL	NUMBER OF ORGANS METASTASIZED	PRESENCE OF CORPORA LUTEA OR LUETINIZED STROMA
							days		
Treated with gonadotropin	A9=56, 8	A8=61 x A8=62	433	++++±	++	++±	130	14	+
	A9=87, 5.5	A8=61 x A8=62	369	++±	++++	++±	149	7	+
	A9=43, 10	A8=66 x A8=62	340	++	+	+	K	5	+
	A9=63, 7.8	A8=59 x A8=64	299	++	++	++	K	12	Corpora hemorrhagica
	A8=20, 13	A7=15 x A7=1	528	+++++	++±	++±	158	6	+
	A7=24, 23	A6=6 x A6=18	1,233	+++	++++	++±	84	8	+
	A7=39, 11.5	A6=22 x A6=27	416	++±	++++	++	99	8	+
	Average....		517	3	2.8	2.1	126	8.6	
Control	A9=54, 8	A8=61 x A8=62	85	+	±	±	K	6	0
	A9=57, 10	A8=61 x A8=62	306	+	+	±	K	3	0
	A9=40, 10	A8=66 x A8=62	176	+++	++	++	172	6	0
	A9=44, 10	A8=66 x A8=62	213	0	±	0	K	1	0
	A9=61, 7.8	A8=59 x A8=64	180	++++	++	++	133	5	0
	A8=38, 10	A7=15 x A7=1	475	+	++	0	204	4	0
	A7=29, 17	A6=22 x A6=18	186	+++	+++	+	160	5	0
	A8=32, 10.3	A7=3 x A6=27	62	+++	+++	+++	95	4	0
	A8=22, 13	A7=13 x A7=1	338	++±	++±	++	134	9	0
	A8=45, 8.6	A7=13 x A7=25	62	+±	+±	±	204	4	0
	Average....		208	2	1.8	1.1	157	4.7	0

that in the controls listed in the table. Likewise, ovariectomy exercised no consistent effect on genetically closely related animals. As these two procedures exercised no effect on the tuberculous process, the data are not listed in detail.

Discussion

It has been demonstrated above that in mature, highly inbred litter mates the synthetic preparation of the naturally occurring estrogenic hormone, α estradiol dipropionate, uniformly retarded the tuberculous process at the site of inoculation in the skin, diminished the extent of the disease in the internal organs, and suppressed to a considerable degree its dissemination in the body.

In rabbits under the age of 4.8 months estrogen exercised a less constant effect and in some instances the tuberculous process was intensified in the internal organs.

By contrast, in a small series of highly inbred mature rabbits, the periodic administration of chorionic gonadotropin, which induced successive crops of corpora lutea in the ovary in the early phase of the disease, uniformly enhanced the tuberculous process at the site of cutaneous inoculation, increased its dissemination, and in the majority of instances aggravated the extent of the disease in the internal organs. Although the number of animals involved was small, the effect appeared specific, for ovariectomy or the daily administration of physiologic quantities of progesterone with several γ of γ estradiol to genetically similar animals exercised no effect on the process. The lack of complete uniformity in the influence of chorionic gonadotropin on the tuberculosis may be related to the inconstant development of fully formed corpora lutea in the ovary throughout the course of the disease in response to the repeated gonadotropic stimulations. As no rabbit gonadotropin has been isolated and, perforce, the human protein was used, it may not be surprising that occasionally the sera of the rabbits chronically treated with the gonadotropin inactivated the hormone, as had been observed by others (8).

Because of this inherent difficulty no further studies were attempted. Nevertheless, the results strongly suggest that gonadotropin affects the tuberculous process in a manner opposite to that of estrogen.

The observed effect of estrogen on the tuberculous process is in accord with the studies of Sprunt and McDearman (9), Foley and Aycock (10) and von Haam and Rosenfeld (11), who have shown that estrogen exerts a protective influence against acute infections caused by vaccinia virus, Streptococcus and Pneumococcus, respectively. However, investigators of the effect of estrogen in the chronic infection, tuberculosis, are quite contradictory in their conclusions. Some assert that it enhances the disease (12), others hold that it exerts no effect on the process (13). Similar differences of opinion have been expressed on the effect of gonadotropin on tuberculosis (14). These varying conclusions suggest that the effects cannot be of great magnitude. In the present experiments the factor of the natural resistance of the control and experimental animals was carefully balanced by the use of highly inbred litter mates of genetically uniform stock and of closely similar hereditary resistance to the disease (1). Under such conditions it should be possible to observe an effect which might be masked by natural variations in resistance of genetically differing animals. This was indeed the case. Furthermore, it has been shown by Sprunt and confirmed in the present studies that estrogen retards the spread of particles in the skin. For this and other reasons the cutaneous portal of entry was used. As it has been shown by Thomas and Duran-Reynals (15) that the incorporation of hyaluronidase in the oculum of tubercle bacilli injected intracutaneously enhances the disease, it is to be expected that a natural hormone which increases the connective tissue permeability to particulate matter would also accelerate the tuberculous process.

As will be shown in subsequent papers, however, mere reduction in spread is not the only effect exercised by estrogen on the rabbit and hence may not be the only effective mechanism by which it retards the disease.

To what extent these data apply to human tuberculosis is problematical. It is well known that the incidence of tuberculous infection as distinguished from clinically significant tuberculous disease increases regularly with age. However, the mortality and morbidity of the disease rises disproportionately synchronous with the onset of puberty, particularly in the female. This sudden change can be understood on the hypothesis that the sex hormones exercise the same effects in man and rabbits. With the onset of the menstrual cycle, a latent tuberculous focus is alternately under the influence of estrogen in the first portion of the cycle and under the corpus luteum, for some time preceding menstruation, in the latter half. It is conceivable that estrogen retards the spread of the disease in the first portion of the cycle, as it does in rabbits, whereas in the latter half the reduction of estrogen, together with the activity of the corpus luteum, induces a spurt of spread in the heretofore latent focus in the same manner as the corpora lutea induced by gonadotropin appear to increase the dissemination of the disease in the animal. However, as the estrogen used in these experiments far exceeds the physiological levels, it is questionable whether these observations can be directly applied to the human disease.

Summary

In highly inbred, sexually mature rabbits estrogen in large doses uniformly retarded the progress of the disease at the site of intracutaneous inoculation, diminished the extent of the disease in the internal organs, and suppressed to a considerable degree its dissemination in the body. In immature rabbits the estrogen was less effective on the spread of the disease. In a smaller group of highly inbred rabbits the periodic administration of chorionic gonadotropin, which induced corpora lutea in the ovary in the earlier phase of the disease, uniformly enhanced the progress of the disease at the site of intracutaneous inoculation, increased its dissemination in the body, and in the majority of instances aggravated its extent in the internal organs. Ovariectomy or the daily combined injection of physiologic amounts of progesterone and estradiol exercised no effect. The possible significance of these hormone effects in the pathogenesis of puberty tuberculosis in the human female is discussed.

Sumario

Factores Constitucionales en la Resistencia a la Infección: I Efecto del Estrógeno y de la Gonadotropina Coriónica sobre la Evolución de la Tuberculosis

En los conejos sexualmente maduros y muy entrecruzados, el estrógeno a grandes dosis retardó el proceso tuberculoso en el sitio de la inoculación intracutánea, disminuyó la extensión de la enfermedad en los órganos internos y suprimió considerablemente su difusión en el cuerpo. En los conejos inmaduros el estrógeno mostró menos efecto sobre la difusión. En un grupo más pequeño de conejos muy entrecruzados, la administración periódica de gonado-

tropina coriónica, que provocó la aparición de cuerpos amarillos en el ovario en la fase más temprana de la dolencia, intensificó invariablemente la agravación de la enfermedad en el sitio de la inoculación intracutánea, acrecentó la difusión en el cuerpo y, en la mayoría de los casos, agravó su extensión en los órganos internos. Ni la ovariectomía ni la inyección combinada a diario de cantidades fisiológicas de progesterona y estradiol ejercieron efecto alguno. Discútese la posible importancia de estos efectos hormónicos sobre la patogenia de la tuberculosis púber en la hembra humana.

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CONSTITUTIONAL FACTORS IN RESISTANCE TO INFECTION[1,2]

II. The Effect of Estrogen on Tuberculin Skin Sensitivity and on the Allergy of the Internal Tissues

MAX B. LURIE, T. N. HARRIS, SAMUEL ABRAMSON[3] AND J. MARVIN ALLISON

Introduction

It was demonstrated in the preceding paper (1) that in mature highly inbred rabbits of genetically similar resistance to tuberculosis, estrogen uniformly retarded the progress of the disease at the site of intracutaneous inoculation and diminished its extent and dissemination in the internal organs. These observations may be understood in the light of the work of Sprunt and his collaborators (2) who found that estrogen reduced the spread of particulate matter in the skin and enhanced the resistance of rabbits to vaccinia virus. Conversely, Thomas and Duran-Reynals (3) have shown that by increasing the penetration of the infecting microorganisms at the portal of entry by the use of a testicular spreading factor, presumably hyaluronidase, the tuberculous process is enhanced not only at the site of inoculation but also in the entire body. Similarly it was found in the preceding study (1) that the periodic injection of chorionic gonadotropin, which engenders successive crops of corpora lutea in the ovary, increases the connective tissue permeability to particles and likewise tends to accelerate the progress of the disease.

While there is a certain degree of parallelism in the present studies between variations in tissue permeability and corresponding differences in the restriction of the disease in the connective tissues, this is far from complete. Hence, further studies were undertaken to elucidate other mechanisms by which estrogen might increase resistance to tuberculosis. As allergic sensitivity is one of the central phenomena in this disease, studies were undertaken to determine the effect of estrogen on the sensitivity of the skin to tuberculin and on the concomitant allergy of the internal organs and tissues.

Materials and Methods

The various groups of estrogen-treated and control rabbits infected with virulent tubercle bacilli, which were described in the previous paper (1), were injected intracutaneously with 0.1 cc. of a 1:10 dilution of the same Old Tuberculin preparation at weekly intervals throughout the course of their disease. The volume of inflammation that developed two days after the injection was determined according to a formula previously given (4) and served as a measure of the degree of skin sensitivity. Similar tests were made on tuberculous rabbits under the respective influence of chorionic gonadotropin, physiologic amounts of progesterone and estradiol, and ovariectomy.

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[2] Aided by grants from the Commonwealth Fund and the Life Insurance Medical Research Fund.

[3] Tuberculosis Control Division, Public Health Service, Federal Security Agency.

Estrogen-treated rabbits and controls were also sensitized by the intracutaneous injection of identical amounts of heat-killed bovine tubercle bacilli at the same intervals. This was done to determine the effect of this hormone on skin sensitivity in the absence of active disease. As heretofore, 0.5 mg. of α-estradiol dipropionate in 0.5 cc. of sesame oil was given to the experimental rabbits at weekly intervals for fourteen days before the beginning of sensitization and throughout the study thereafter. The control animals received 0.5 cc. of sesame oil without the hormone at the same time.

Since the tuberculin sensitivity induced by treatment with heat-killed tubercle bacilli lasts for many months, and, furthermore, since injected estrogen is eliminated from the body, the effect on skin allergy of withdrawing the hormone and subsequently readministering it was studied.

To determine whether estrogen has the same effect on the sensitivity of the skin and on that of the internal tissues, control and estrogen-treated rabbits, sensitized with heat-killed tubercle bacilli, were injected with 0.2 cc. of a 1:10 dilution of tuberculin subcutaneously instead of intracutaneously. The response was measured by the rise in the absolute number of circulating polymorphonuclear leucocytes. For the same purpose, the extent of caseation in the tuberculous tissues of the estrogen-treated and control animals at death was correlated with the tuberculin skin sensitivity during the animal's life. Tissue culture studies on the effect of tuberculin on white blood cells derived from estrogen-treated and control rabbits, all sensitized with heat-killed tubercle bacilli, were also performed with the same object in view. Methods used in these experiments will be described in detail later. Finally, it became apparent during the course of this work that estrogen might affect the inflammation which a particular agent induced in the tissues apart from the phenomena of sensitization. Hence, several toxic substances were injected into the skin of unsensitized estrogen-treated and control animals, and the volume of the ensuing inflammation was measured as heretofore.

Results

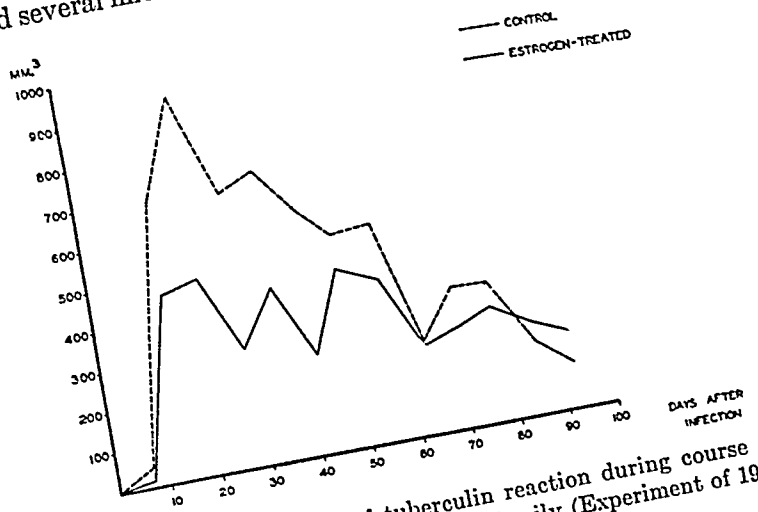
THE EFFECT OF ESTROGEN, CHORIONIC GONADOTROPIN, PROGESTERONE AND OVARIECTOMY ON THE TUBERCULIN SKIN SENSITIVITY OF RABBITS WITH PROGRESSIVE TUBERCULOSIS

Text figure 1 presents the average skin sensitivity in 4 estrogen-treated and 4 litter mate controls of the C family throughout the course of their disease. All the relevant data for this experiment of 1945–1946 are detailed in text figure 3 and table 2 of the preceding paper (1). It is evident that estrogen markedly and consistently reduced the skin sensitivity to tuberculin until very late in the course of the infection, at which time the less extensively diseased hormone-treated animals showed a somewhat higher sensitivity than their controls who were on the verge of death. As is well known, tuberculin skin sensitivity declines markedly during the last stages of tuberculosis.

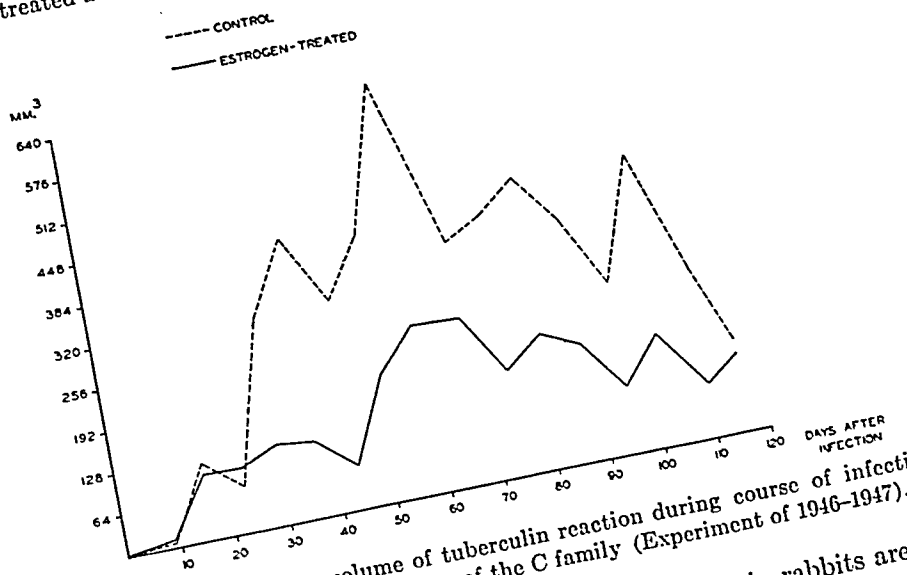
Text figures 2 and 3 present similar observations in the experiments of 1946–1947 on rabbits of the C and A families, respectively, which are detailed in text figures 4 and 5 and tables 3 and 4 of the preceding study (1). It is again clear that estrogen uniformly reduced the skin sensitivity in the same manner as in the experiment of 1945–1946.

Text figure 4 shows the average degree of tuberculin sensitivity of the skin at different intervals during the course of the disease in 5 control rabbits of the

Family A, and in 8 litter mates, 4 of which were under the influence of chorionic gonadotropin and 4 under daily treatment with physiologic quantities of progesterone and several micrograms of estradiol. The exact conditions under which

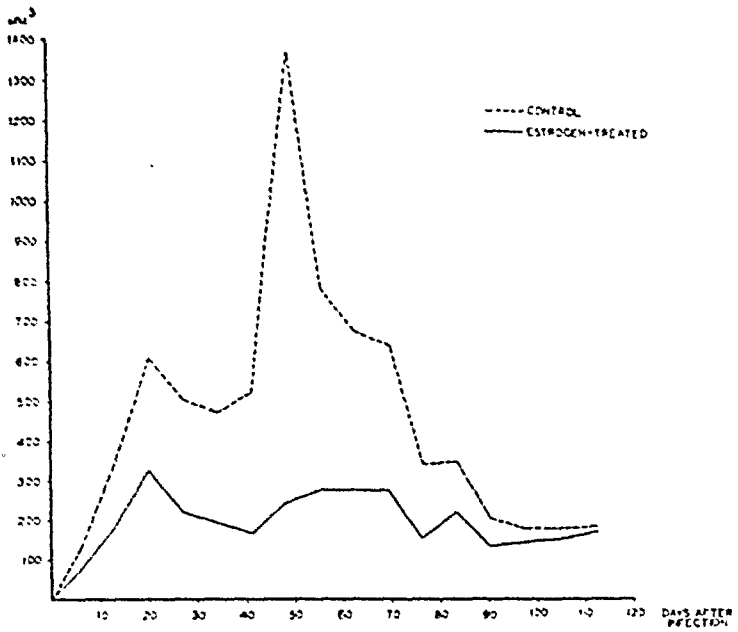


TEXT FIGURE 1. Average volume of tuberculin reaction during course of infection in estrogen-treated and control litter mates of the C family (Experiment of 1945–1946).

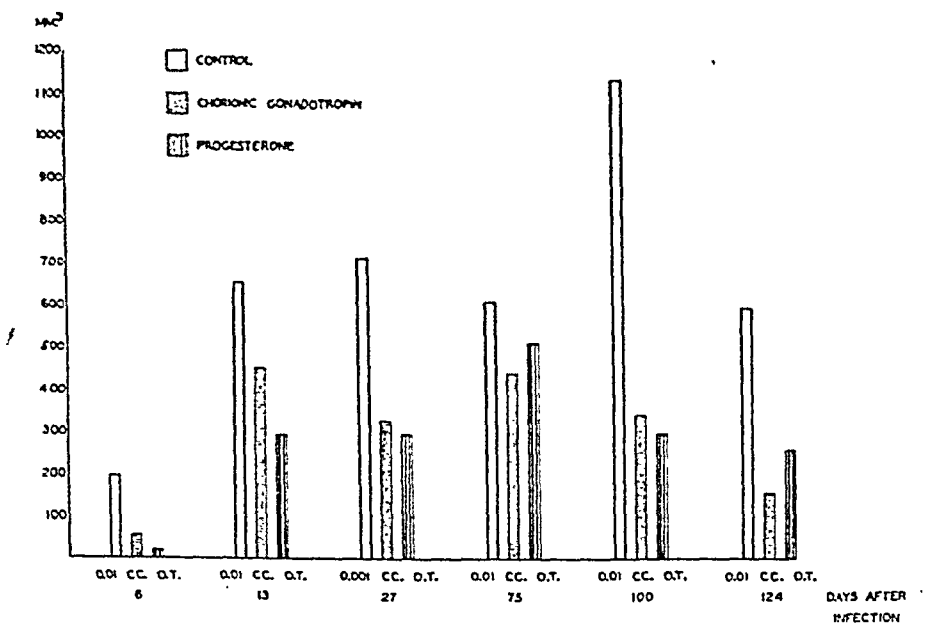


TEXT FIGURE 2. Average volume of tuberculin reaction during course of infection in estrogen-treated and control litter mates of the C family (Experiment of 1946–1947).

these data were obtained from the control and gonadotropin rabbits are given in text figure 6 and table 6 of the preceding report. It is evident that treatment of rabbits with chorionic gonadotropin, which it will be remembered (1) tended to enhance the disease, and progesterone-estrogen injections, which did not ma-



TEXT FIGURE 3. Average volume of tuberculin reaction during course of infection in estrogen-treated and control litter mates of the A family (Experiment of 1946–1947).



TEXT FIGURE 4. Average volume of tuberculin reaction during course of infection in chorionic gonadotropin-treated, progesterone-treated and control litter mates of the A family (Experiment of 1944–1945).

terially affect the process, both consistently lowered the skin sensitivity as compared with that observed in their litter mate controls. Furthermore, ovar-

iectomy, which was found to exercise no discernible influence on the progress of the disease, also lowered skin sensitivity. It is apparent, therefore, that the degree of tuberculin skin sensitivity has no constant relation to the progress of the disease. Hence, the observed retardation of the tuberculosis in the estrogen-treated litter mates is not accounted for by the depression of the skin sensitivity which follows treatment by this hormone.

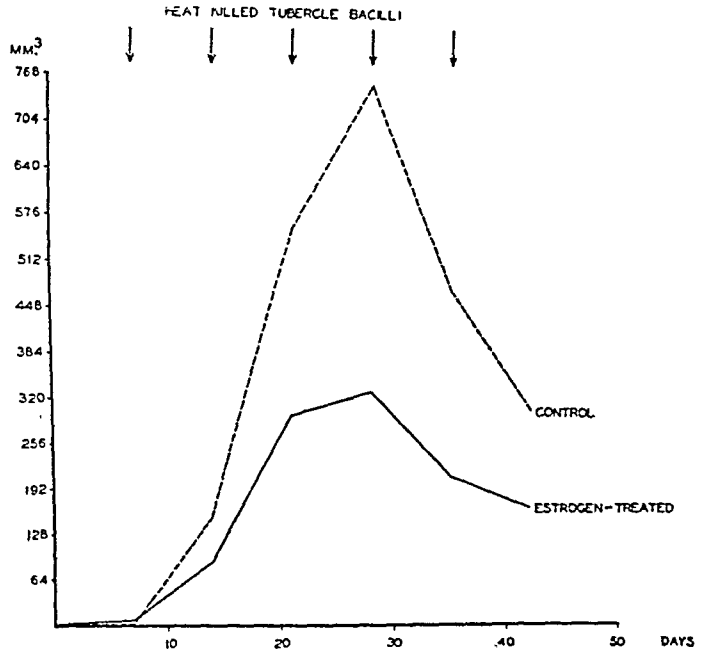
THE EFFECT OF ESTROGEN ON THE TUBERCULIN SKIN SENSITIVITY OF RABBITS SENSITIZED WITH HEAT-KILLED TUBERCLE BACILLI

While there is no strict parallelism between the extent of the disease and the intensity of skin allergy, one may still contend that, *ceteris paribus*, the less marked the antigenic stimulus the less will be the allergy. Since estrogen retarded the tuberculosis, it might be presumed that one factor in this reduction of allergy is the less extensive disease of the estrogen-treated animals. That this is not the case is illustrated in text figure 5 which presents the average degree of tuberculin sensitivity in 5 estrogen-treated and 5 control genetically closely related rabbits on each of six successive weeks. All of these rabbits had been given the same quantities of heat-killed tubercle bacilli intracutaneously at the same intervals as indicated in the figure. It is evident, therefore, that identical nonmultiplying antigenic stimuli result in a lower skin sensitivity in estrogen-treated than in control animals.

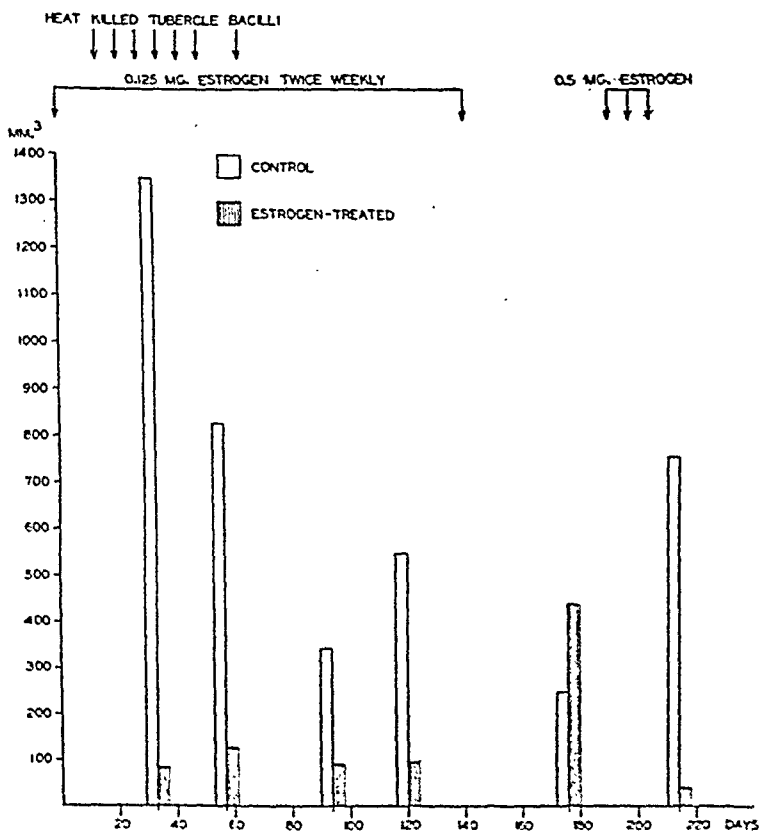
THE EFFECT OF WITHDRAWAL AND READMINISTRATION OF ESTROGEN ON TUBERCULIN SKIN ALLERGY IN RABBITS SENSITIZED WITH HEAT-KILLED TUBERCLE BACILLI

The depressed skin allergy to tuberculin in estrogen-treated rabbits sensitized by active tuberculosis or by treatment with heat-killed tubercle bacilli may be due to a fundamental depression of the sensitizing mechanism or it may simply result from an alteration of the inflammatory irritability of the skin. That the second is the more likely explanation may be seen from text figure 6.

Three estrogen-treated and three control rabbits were sensitized with heat-killed tubercle bacilli as indicated in the figure. For the first 140 days the rabbits received 0.125 mg. α-estradiol dipropionate twice weekly. During this time both groups were repeatedly tested with tuberclin. In each instance the sensitivity of the rabbits under the influence of the hormone was consistently lower than that of the controls. The hormone was now withdrawn and thirty-six days after the last injection of estrogen the rabbits were again tested with tuberculin. As is evident from the figure, the sensitivity was reversed and the rabbits, whose skin allergy had been depressed during the estrogen administration, now reacted with even greater intensity than the control animals. The experimental rabbits were then again given estrogen for three weeks. Thirty-six days after the second hormone withdrawal, the sensitivity was again restored. It is clear, therefore, that the lowered sensitivity of the skin in rabbits under the influence of estrogen is not due to a depression of the level of skin sensitization,



TEXT FIGURE 5. Average volume of tuberculin reaction after sensitization with heat-killed tubercle bacilli in estrogen-treated and control inbred rabbits (Experiment of 1946).



TEXT FIGURE 6. The effect of estrogen and its withdrawal and readministration on tuberculin skin allergy in rabbits sensitized with heat-killed tubercle bacilli (Experiment of 1946–1947).

but to some effect of the hormone on the inflammatory response of the skin to tuberculin.

THE EFFECT OF ESTROGEN ON THE ALLERGIC SENSITIVITY OF THE INTERNAL TISSUES

The question now arose whether the body as a whole shares with the skin in the depression of the inflammatory responsiveness to the products of the tubercle bacillus as a result of estrogen administration. It was found, in the first place, that the extent of caseation in the tuberculous foci was in general at least as pronounced in the estrogen-treated animals as in their litter mate controls. Furthermore, in the former group, exudation into the alveoli of the lungs with necrosis was prominent in the advancing portion of the lesion, whereas in the latter animals, thickening of the alveolar septa by a productive inflammation was most marked and caseation was less widespread. If caseation is mediated

TABLE 1

The effect of estrogen on the rise of polymorphonuclear counts in tuberculin-sensitive rabbits induced by the subcutaneous injection of tuberculin

GROUP	RABBIT NUMBER	ORIGINAL NUMBER OF POLYMORPHONUCLEARS	NUMBER OF POLYMORPHONUCLEARS 24 HOURS AFTER TUBERCULIN	RISE IN POLYMORPHONUCLEARS	GROUP	RABBIT NUMBER	ORIGINAL NUMBER OF POLYMORPHONUCLEARS	NUMBER OF POLYMORPHONUCLEARS 24 HOURS AFTER TUBERCULIN	RISE IN POLYMORPHONUCLEARS
				per cent					*per cent*
Untreated controls	V 414	5,119	7,330	43.2	Estrogen-treated	V 267	2,936	7,712	162.6
	V 366	2,983	6,763	126.7		V 30	1,785	5,418	203.5
	A 6	1,760	7,893	348.4		A 5	1,625	4,956	204.8
	A 9	1,534	6,767	341.1		A 8	1,468	5,763	292.5
	A 11	2,303	5,486	138.2					
	Average...	2,740	6,848	199.5		Average...	1,953	5,962	215.8

by allergic sensitivity, for which there is some evidence, then estrogen does not reduce this sensitivity in the lungs.

It is difficult, however, to evaluate the extent of caseation in terms of allergic sensitivity if the complexity of the factors involved are considered. Therefore, in an effort to bypass the skin, control and estrogen-treated rabbits which had been sensitized with heat-killed tubercle bacilli were given a subcutaneous injection of 0.2 cc. of 1:10 dilution in saline of Old Tuberculin on Long's synthetic medium. The response was measured by the absolute rise in circulating polymorphonuclear leukocytes on the day following injection.

Table 1 shows the results of such an experiment. It is evident that sensitized rabbits, whether under the influence of estrogen or not, reacted to the subcutaneous injection of tuberculin with a sharp rise in polymorphonuclear leukocytes which is of the same degree in both groups. It is noteworthy that sterile Long's synthetic medium concentrated on the waterbath to one-tenth its original volume, *i.e.*, control material, caused no significant change in the leukocyte count

on subcutaneous injection. It is thus apparent that estrogen did not lower the leukocytic response of allergic rabbits to tuberculin.

THE TUBERCULIN SENSITIVITY OF EXPLANTS OF BLOOD LEUKOCYTES DERIVED FROM SENSITIZED, ESTROGEN-TREATED AND CONTROL RABBITS

Since the original tissue culture studies of Rich and Lewis (5) on the tuberculin reaction have shed the clearest light on the phenomenon, an attempt was made to determine whether any difference in cell sensitivity to tuberculin could be demonstrated in explants of blood leukocytes derived from estrogen-treated and control rabbits, sensitized with heat-killed tubercle bacilli.

Three estrogen-treated and three control rabbits were sensitized with heat-killed tubercle bacilli as detailed above in text figure 6. After repeated tuberculin skin tests had shown that in the rabbits under the influence of the hormone the inflammatory response was markedly depressed, blood was withdrawn from the heart of the rabbits of both groups. The citrated blood was centrifuged and the leucocytic layer was coagulated with 0.5 per cent calcium chloride in Gey's solution (6). The tissue was minced into pieces of approximately 1.0 mm. in diameter, washed with Gey's solution, and picked up by a Pasteur pipet bent at the tip and placed in a test tube 150 by 18 mm. The arrangement of the explants was as follows. Five bits of tissue from an experimental animal were placed in a straight line extending approximately halfway up the tube. A few drops of the clotting mixture were then run down the tube along the line of explants and allowed to remain in a horizontal position until coagulation had occurred. The same procedure was then repeated with five bits of tissue from a control animal along the opposite side of the same tube. The clotting mixture consisted of equal amounts of heparinized chicken plasma, and 10 day chick embryo extract in Gey's solution. The latter contained the appropriate dilution of Old Tuberculin prepared on Long's synthetic medium, or the concentrated sterile medium which served as control for the tuberculin. In the case of each row of explants the tube was manipulated so that each explant was surrounded by several millimeters of clot. The tubes were then stoppered and incubated at 37°C in a roller-tube mechanism. The cultures were examined after twenty-four hours and the farthest distance of migration of leucocytes from the periphery of the explant was measured by an ocular micrometer. These radii were determined in four directions at 90 degree intervals around the explant, and the mean was taken as the average distance of migration. The results are given in arbitrary units of the ocular micrometer used in these experiments.

In preliminary experiments it was found that the preparation of tuberculin used in these experiments showed a very marked inhibition of migration of the leukocytes of sensitized rabbits at a concentration of 1:500 and no statistically significant effect at 1:4,000. In order to allow an opportunity for the demonstration of any difference in sensitivity between estrogen-treated and control rabbits it was thought best to use the intermediate concentration of 1:1,000.

It was observed that a 1:1,000 solution of uninoculated medium showed no effect on the tissue cultures. Hence it was considered justifiable to attribute the inhibitory effect of tuberculin at concentrations of 1:1,000 to specific sensitization and to use saline solution as control material for the tuberculin.

In the major series of tissue culture experiments, ten explants of leukocytes from each rabbit were cultivated in the presence of tuberculin at concentrations

of 1:1,000, 1:4,000, and zero. The mean radius of migration of leucocytes for each of the 10 specimens is shown in table 2.

It will be seen that in all explants derived from both estrogen-treated and control animals there was a very small difference between cultures grown in saline and those grown in 1:4,000 tuberculin. In no instance, however, was the difference statistically significant. In contrast, the cultures grown in a medium containing tuberculin in a concentration of 1:1,000 all showed a marked decrease in radius of migration as compared with the control cultures. Analysis of the data by the Student method revealed this difference to be statistically significant.

A comparison of the effect of this concentration of tuberculin (1:1,000) on the migration of leukocytes derived from estrogen-treated and control animals showed that explants from two of the estrogen-treated rabbits were inhibited

TABLE 2

Average radius of migration in tissue culture of leukocytes derived from sensitized estrogen-treated and control rabbits in the presence of various concentrations of tuberculin together with the corresponding sensitivity of the skin to tuberculin†*

TREATED WITH ESTROGEN						CONTROL, UNTREATED					
Rabbit Number	Radius of migration in salt solution	Radius of migration in 1:4,000 tuberculin	Radius of migration in 1:1,000 tuberculin	Reduction of migration in 1:1,000 tuberculin	Skin sensitivity of inflammation	Rabbit Number	Radius of migration in salt solution	Radius of migration in 1:4,000 tuberculin	Radius of migration in 1:1,000 tuberculin	Reduction of migration in 1:1,000 tuberculin	Skin sensitivity of inflammation
				per cent	*mm.³***					*per cent*	*mm.³***
A 5	341	319	218	36.1	84	A 6	309	321	230	25.6	637
A 8	385	350	203	47.4	153	A 9	299	323	216	27.8	323
A 10	320	291	201	37.2	45	A 11	268	242	161	39.9	383

* Each value is an average of 10 explants expressed in arbitrary micrometer units.
† Represents the average value of inflammation to 0.1 cc. of 1:10 dilution of tuberculin two weeks before and four days after the leukocytes were cultured.
** Per cubic millimeter of free flowing, venous blood.

in their migration by tuberculin to a somewhat greater extent than those derived from two untreated sensitized controls. The explants from the remaining experimental and control rabbits were equally inhibited by this concentration of tuberculin. These differences, however, are not statistically significant.

By contrast, the skin sensitivity to tuberculin of each rabbit under influence of estrogen, tested two weeks before and four days after the tissue cultures were done, was markedly depressed in comparison with that of the sensitized controls.

It is evident, therefore, that tissue culture also failed to show any difference in the degree of cell sensitivity to tuberculin as a result of estrogen treatment.

THE INFLAMMATORY RESPONSE OF UNSENSITIZED ESTROGEN-TREATED AND CONTROL RABBITS TO BACTERIAL IRRITANTS

It is apparent from the work thus far reported that there is no evidence that estrogen depresses the allergic irritability of the internal tissues of sensitized

rabbits. Even what appears to be a reduction in the tuberculin sensitivity of the skin is not due to a lower degree of sensitization of this tissue, but rather to an alteration induced by the hormone in the inflammatory responsiveness of the skin to the irritant. That the intrinsic allergic irritability was unaffected was demonstrated by the withdrawal of the hormone.

TABLE 3

The effect of estrogen on the inflammatory irritability of the skin to heat-killed tubercle bacilli and to pertussis endotoxin

RESPONSE TO HEAT-KILLED TUBERCLE BACILLI			RESPONSE TO PERTUSSIS ENDOTOXIN		
Group	Rabbit number	Volume of inflammation	Group	Rabbit number	Volume of inflammation
		*mm.*³*			*mm.*³*
Treated with estrogen	V 30	44	Treated with estrogen	V 30	76
	V 366	34		V 366	81
	V 784	54		V 784	144
	V 267	26		V 267	28
	V 414	39		V 414	50
	Average.....	39		Average.....	76
Untreated controls	T 33	69	Untreated controls	T 33	68
	V 685	140		V 685	392
	V 368	85		V 368	432
	U 57	76		U 57	118
	S 376	97		S 376	54
	Average.....	93		Average.....	213

* Per cubic millimeter of free flowing, venous blood.

That this is the correct explanation for the reduction of the skin sensitivity by estrogen may be seen from the following observations.

Ten unsensitized genetically closely related rabbits were divided into two groups. Five were treated with estrogen as usual. The remaining animals served as controls. After the experimental animals had been under the influence of estrogen for twenty days, both groups received 1.0 mg. of heat-killed tubercle bacilli of the Ravenel strain intracutaneously. The extent of the inflammation at the site of injection was measured in both groups forty-eight hours later. On another occasion both groups were injected intracutaneously with 0.1 cc. of 1:200 dilution in saline of a sonic vibration extract of *Bacillus pertussis*[4] (7). On the second day the ensuing inflammation was measured. The results are presented in table 3.

It is evident that the inflammatory reaction to both bacterial agents is reduced in the skin of estrogen-treated rabbits. As the rabbits received these irritants for the first time, no phenomena of sensitization were involved. Thus two substances of primary toxicity to the skin induced less inflammation in animals under

[4] The pertussis endotoxin was kindly furnished by Dr. Joseph Smolens.

the influence of the hormone than in control rabbits. Therefore, the depression of the tuberculin skin reaction in sensitized rabbits treated with estrogen is due to a lower inflammatory responsiveness of the skin to bacterial irritants in general.

A similar depression of the inflammatory responsiveness of the skin was noted in a study of the Arthus phenomenon in estrogen-treated and control rabbits sensitized with a protein of the tubercle bacillus kindly furnished by Dr. F. B. Seibert (8). After sensitization to the protein had been achieved, and in the presence of approximately equal precipitin titres in the serum of both groups, the inflammation at the site of intracutaneous injection of the antigen was usually much less intense in the estrogen-treated animals.

Discussion

The inflammatory response to tuberculin of the skin of rabbits sensitized by active tuberculosis or by treatment with heat-killed tubercle bacilli is markedly depressed by the administration of estrogen. At the same time, however, the polymorphonuclear leukocytosis which follows the subcutaneous administration of tuberculin in sensitized rabbits is equally intense whether the rabbits are under the influence of the hormone or not. Similarly, explants of tissue derived from sensitized rabbits, whether estrogen-treated or not, are equally inhibited in their migration in tissue cultures in the presence of the same concentration of tuberculin. Furthermore, the extent of caseation in estrogen-treated rabbits with progressive tuberculosis is not only not reduced but may even be increased. Thus the internal tissues are not depressed in their allergic response to the tubercle bacillus or its products, as is the skin. Even what appears as a reduction of the skin sensitivity to tuberculin is not due to a depression of the acquired degree of sensitivity of the skin, as a result of an interference by estrogen with the sensitizing mechanism. For, on withdrawal of the hormone, the sensitivity of the skin, without any additional sensitization, becomes at least as pronounced as that of control rabbits similarly sensitized. This heightened sensitivity can now again be depressed by further estrogen administration. Estrogen thus masks an existing unaltered allergic irritability. Finally the skin of estrogen-treated rabbits responds with less inflammation to unrelated primary toxic agents given to unsensitized rabbits. Hence the reduction in the skin sensitivity to tuberculin of estrogen-treated rabbits, whether sensitized by heat-killed tubercle bacilli or by active disease, is due not to diminution of skin sensitization, but simply to a depression of the inflammatory responsiveness of the skin to toxic agents in general. Among these agents is tuberculin, which is toxic to sensitized rabbits.

Analysis of the effect of estrogen on the tuberculin sensitivity of the skin has failed to shed any light on the mechanism whereby this hormone retards the tuberculous process. Nevertheless, the studies reveal that under certain conditions the tuberculin reaction of the skin is an unreliable index not only of the degree of sensitization of the internal organs, but even of that of the skin itself. The phenomenon was observed with estrogen which masks an existing allergic irritability by depressing the inflammatory response of the skin to irritants in

general. The importance of this observation in the interpretation of the problem of the relation between allergy and immunity is obvious.

Summary and Conclusions

1. Estrogen reduced the inflammatory response of the skin to tuberculin in rabbits sensitized by active tuberculosis or by treatment with heat-killed tubercle bacilli by virtue of the depressing effect of the hormone on the inflammatory irritability of the skin to bacterial irritants in general.

2. The acquisition of intrinsic allergic sensitivity of the tissues in general, as well as that of the skin, is not reduced by estrogen administration.

Sumario

Factores Constitucionales en la Resistencia a la Infección: II. El Efecto del Estrógeno sobre la Cutisensibilidad a la Tuberculina y sobre la Alergia de los Tejidos Internos

1. El estrógeno atenuó la respuesta inflamatoria de la piel a la tuberculina en los conejos sensibilizados por tuberculosis activa o por tratamiento con bacilos tuberculosos muertos al calor, por virtud del efecto depresor que ejerce la hormona sobre la irritabilidad inflamatoria de la piel a los irritantes bacterianos en general.

2. La adquisición de sensibilidad alérgica intrínseca por los tejidos en general, así como por la piel, no es rebajada por la administración de estrógeno.

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CONSTITUTIONAL FACTORS IN RESISTANCE TO INFECTION[1,2]

III. On the Mode of Action of Estrogen and Gonadotropin on the Progress of Tuberculosis

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Introduction

In the preceding studies (1, 2) an endeavor was made to elucidate the mechanism whereby, on the one hand, estrogen retarded the progress of tuberculosis and, on the other, chorionic gonadotropin enhanced the dissemination of the disease. It was found (2) that estrogen markedly suppresses the inflammatory response of the skin to tuberculin in rabbits sensitized by active tuberculosis or by treatment with heat-killed tubercle bacilli. It was also found, however, that estrogen reduces the inflammatory response of the skin to bacterial toxic agents in general and hence also to tuberculin, which is toxic to sensitized animals. Therefore, this seemingly depressing effect of estrogen on allergy, one of the important elements in the tuberculous process, is not due to an essential alteration in the allergizing mechanism but rather to the masking of an existing unaffected allergy by the hormone. What role, if any, this reduction in the inflammatory response of the skin plays in the retardation of tuberculosis by the hormone is not clear.

In a further effort to demarcate the means by which estrogen retards the progress of tuberculosis, information was sought as to the effect of estrogen on the rate of multiplication of tubercle bacilli at the site of inoculation in the skin and in the metastatic foci. Bacterial infection takes place chiefly in the connective tissue. As the outstanding effect of estrogen on tuberculosis is its restricting influence on the dissemination of the infection, whereas that of gonadotropin is the enhancement of this process, studies on the alterations produced by these hormones in the constituents of the connective tissue were instituted. An endeavor was made to determine whether estrogen affected the concentration of water in the connective tissue of the skin. Studies were also undertaken to determine the effect of estrogen and chorionic gonadotropin on the hyaluronic acid of the skin and on its vascular permeability; for these factors play a significant role in the dissemination of particles and bacteria in the connective tissues.

As stated in the second paper of this series (2), the parallel observed between the effect of estrogen on the spread of particles on the one hand, and on the progress of the disease on the other, was not constant. Therefore, other clues were sought to explain the observations.

[1] From The Henry Phipps Institute, University of Pennsylvania, Philadelphia, Pennsylvania.

[2] Aided by a grant from the Commonwealth Fund.

[3] Tuberculosis Control Division, Public Health Service, Federal Security Agency.

[4] Dorothy Temple Cross Research Fellow of the Medical Research Council of Great Britain.

In view of the well known interrelation between the gonads and the adrenals and the studies of Selye (3) on the adaptation syndrome and, in view of the demonstration by White and Dougherty (4) of the controlling effect of the adrenal corticosterones on the lymphocytes and their dissolution, and furthermore in consideration of the observations of McMaster and Hudack (5) and Ehrich and Harris (6), which strongly suggest that lymphocytes play a significant role in antibody production, the effect of the administration of estrogen on the adrenal cortex and on antibody formation was studied. The influence of this hormone on the thyroid gland was also noted.

Finally, during the course of this investigation, it was found that estrogen markedly suppressed the usually extensive amyloid degeneration in the spleen of rabbits with chronic tuberculosis. In the following pages these data are evaluated and an attempt is made to integrate the observations in the light of present knowledge.

TABLE 1

Fate of tubercle bacilli at the portal of entry and in the draining nodes and kidneys of estrogen-treated and normal rabbits

Rabbit number		Days after infection	Number of viable tubercle bacilli on culture					
			Local lesion		Draining nodes		Kidney	
Experimental; age in months at time of infection	Control; age in months at time of infection		Experimental	Control	Experimental	Control	Experimental	Control
A9=98; 17	A9=99; 17	2	80	624	750	1,225	1	1
A9=102; 17	A9=103; 17	7	1,140	1,116	29,800	14,520	0	0
A9=135; 4	A9=138; 4	14	56,000	43,000	260,000	—	0	?
A10=7; 4	A10=10; 4	28	11,000	2,000	106,500	133,500	0	0

Observations

THE EFFECT OF ESTROGEN ON THE RATE OF MULTIPLICATION OF TUBERCLE BACILLI IN THE TISSUES

It was reported by Faulkner (7) that the estrogenic substance, diethylstilbestrol, is bactericidal to tubercle bacilli *in vitro*. It became necessary, therefore, to determine whether the natural estrogen, estradiol, used in these studies retarded the tuberculous process by suppressing the multiplication of the bacilli in the tissues. To answer this question, four litter mate pairs of the A family were divided into two groups. One received 0.5 mg. of α-estradiol dipropionate subcutaneously in sesame oil once a week, and the other received the same amount of oil without the hormone. When the experimental animals had been under the influence of estrogen for two weeks, both groups were infected intracutaneously with virulent bovine tubercle bacilli. At various intervals following infection, weighed amounts of the cutaneous lesion, the draining lymph nodes, and the internal organs were cultured on a modified Löwenstein medium (8) and the number of viable tubercle bacilli that they contained was determined as

described in previous studies (9). Estrogen was administered at weekly intervals throughout the course of the experiment.

The results are presented in table 1. It is evident that estrogen exercises no constant effect on the growth of tubercle bacilli at the site of inoculation or in the metastatic foci in the early stages of the infection.

THE EFFECT OF ESTROGEN AND GONADOTROPIN ON THE CONSTITUENTS OF THE CONNECTIVE TISSUE OF THE SKIN

Table 2 shows the effect of estrogen treatment on the spread of India ink in the rabbits of the experiment cited in the above paragraph, previous to infection. Included in this table are similar data on a group of litter mates of the C family. Clearly, estrogen usually restricts the spread of particles in the skin to a considerable degree, as shown previously by Sprunt (10).

TABLE 2

The effect of estrogen on the spread of India ink particles in the skin

GROUP	RABBIT NUMBER	1 DAY SPREAD MM²*	RABBIT NUMBER	1 DAY SPREAD MM²
Untreated controls	A9=99	1,187	CS=2	1,470
	A9=103	1,800	CS=17	2,122
	A9=138	1,458	CS=19	1,850
	A10=10	1,722	CS=23	2,787
	Average.....	1,542	Average	2,057
Estrogen treated	A9=98	567	CS=1	1,120
	A9=102	1,367	CS=16	1,410
	A9=135	1,361	CS=18	1,990
	A10=7	967	CS=24	1,120
	Average.....	1,066	Average	1,410

* Area of spread in square millimeters as determined by planimeter.

THE EFFECT OF ESTROGEN ON THE CONCENTRATION OF WATER IN THE SKIN

As stated above, it is possible that much of the retardation of the progress of tuberculosis induced by estrogen may be due to its restricting effect on the spread of particles, such as tubercle bacilli, in the tissues. Therefore, the mechanism whereby estrogen reduces the permeability of the connective tissue was investigated.

It is well known that estrogen produces a great increase in the water content of the sexual skin of monkeys. Furthermore, it has been demonstrated by Zuckerman (11) that estrogen increases the water of the skin of mice. If rabbits are similarly affected by the hormone, a simple explanation for the restricting effect of estrogen on the spread of particles would be at hand. The water in the connective tissue is not in a free state but is either intracellular or bound to the matrix or in a film about the fibres and cells (12). As a result of the introduction of estrogen, these cells and fibres may swell. Thus the turgescence of the connec-

tive tissue elements would increase and in this way hinder their disruption by particles introduced into their interstices. Accordingly, the water content of the skin of estrogen-treated and control, litter mate rabbits was determined.

About one gram of clipped skin was excised with sharp, curved scissors. After removing the adhering blood with gauze, the skin was quickly placed in a previously accurately weighed covered bottle and heated to a constant weight at 100° C. From these data the concentration of water in the skin of estrogen-treated and control animals was determined. The observations are presented in table 3.

It is evident that estrogen does not increase the total concentration of the water in the skin. Whether the hormone alters the distribution of water between the extra- and intracellular compartments has not been determined.

TABLE 3

The effect of estrogen on the concentration of water in the skin

GROUP	RABBIT NUMBER	PER CENT OF WATER	GROUP	RABBIT NUMBER	PER CENT OF WATER
Untreated controls	T 33	67	Untreated controls	A9=99	63.3
	V 685	69		A9=103	67.9
	V 368	67		A9=138	71.3
	U 57	69		A10=10	75.4
	S 376	65.5			
	Average.....	67.5		Average.....	69.5
Treated with estrogen	V 30	65	Treated with estrogen	A9=98	69.0
	V 366	67		A9=102	68.4
	U 784	68		A9=135	74.2
	V 267	67		A10=7	68.5
	V 414	69.7			
	Average.....	67.3		Average.....	70.0

THE EFFECT OF ESTROGEN AND CHORIONIC GONADOTROPIN ON THE SPREADING INFLUENCE OF HYALURONIDASE

It was shown in a previous study (13) that chorionic gonadotropin enhances the spread of India ink in the skin. The process is mediated by the corpora lutea engendered in the ovary by the gonadotropin for in ovariectomized animals this hormone has no such effect. Furthermore, progesterone also tends to enhance the spread of dyes in the skin. Estrogen exercises an opposite effect, restricting the spread of particles. Hence it was thought possible in connection with the permeability of the skin, that the interrelation between the enzyme hyaluronidase and its substrate hyaluronic acid may be under the influence of the sex hormones (14). Further support for this notion is afforded by the fact that a substance similar to hyaluronic acid accumulates (15) in the sex skin of monkeys, under the influence of estrogen.

Accordingly 3 litter mates of the tenth inbred generation of the A family were divided into three classes. Five such litter mate groups were thus divided. The first class of 5 rabbits received 0.5 mg. α-estradiol dipropionate subcutaneously in 0.5 cc. sesame oil at weekly intervals for a total period of three weeks. The second class of 5 rabbits served as controls and received the same weekly injection of oil without the hormone. The last 5 rabbits received 0.06 mg. of chorionic gonadotropin intravenously at ten day intervals. This amount of gonadotropin regularly engendered crops of corpora lutea in the ovary.

Six days after the last injection of estrogen and two days after the last treatment with chorionic gonadotropin, the spreading effect of hyaluronidase[5] on India ink and on rabbit hemoglobin injected in the skin of the above three groups of litter mates was measured. It was believed that, if the character or concentration of hyaluronic acid in the skin of animals under the influence of these two hormones differs, the same amount of hyaluronidase would exercise a differential spreading effect in animals subjected to the respective hormone.

Three tenths cc. of saline containing the same amount of India ink or hemoglobin without hyaluronidase, and the same volume and concentration of the two dyes containing 1 and 5 γ of the enzyme per cc., respectively, were injected into corresponding areas of each of the 15 rabbits in the three above groups. The spread of these two dyes in the six sites of each of these rabbits was measured by calipers fifteen minutes after injection. The area of spread was determined as suggested by Hechter (16).

An examination of table 4, which summarizes these observations, reveals that under all conditions, with or without hyaluronidase, the spread of India ink particles or of diffusible rabbit hemoglobin tends to be restricted in the estrogen-treated rabbits and enhanced in rabbits under the influence of gonadotropin as compared with that in untreated litter mates. This confirms the original observations (13).

In general, 5 γ of hyaluronidase per cc. enhanced the spread of both dyes to a greater extent than 1 γ. Apparently, therefore, the enhancement of spread by hyaluronidase is proportional to its concentration. Furthermore, hyaluronidase has a much greater enhancing effect on the spread of diffusible hemoglobin than on that of particulate India ink. As tubercle bacilli are particulate, it is evident that, from the standpoint of the mechanism of action of these hormones on the tuberculous process, the effect of hyaluronidase on the spread of particulate ink in rabbits under the influence of these two hormones is of greater significance than the effect of this enzyme on diffusible hemoglobin.

If attention is concentrated on the effect of both doses of hyaluronidase on the spread of India ink in estrogen-treated rabbits and in litter mates under the influence of gonadotropin, it is evident that the enzyme enhances the spread of particles to a greater extent in the former than in the latter. This difference, though only 20 per cent in extent, is, nevertheless, statistically significant, for the critical ratio of the difference is 3.3 with a "P" value of 0.002. Essentially

[5] Highly purified sheep testicle hyaluronidase (VIII 12S) was kindly placed at our disposal by Dr. Karl Meyer.

the same diminution in the spreading effect of hyaluronidase on hemoglobin was seen in animals under the influence of gonadotropin as distinguished from those under the effects of estrogen. In this case, however, the difference was not quite statistically significant for the "P" value was 0.04. Furthermore, hyaluronidase enhanced the spread of India ink to a slightly greater extent in estrogen-treated rabbits than in normal litter mates. The difference was observed in only half of the tests, however, and the results were not statistically significant.

The enhancement of spread induced by hyaluronidase is due to the hydrolysis of the hyaluronic acid matrix of the connective tissue (14). Therefore, the enzyme depolymerizes this intercellular substance over a wider area or more completely in estrogen-treated rabbits than in animals under the influence of gonadotropin. A similar though less constant difference would appear to obtain

TABLE 4

The effect of hyaluronidase on the spread of India ink in the skin of normal, estrogen-treated, and gonadotropin-treated litter mates of the inbred rabbit family A

GROUP	NUMBER OF RABBITS	SPREAD OF INDIA INK IN MM²					SPREAD OF HEMOGLOBIN IN MM²		
		Without hyaluronidase	With hyaluronidase		Average of 10 observations	Increment of spread for both doses	Without hyaluronidase	With 5 γ of hyaluronidase per cc.	Increment of spread
			1 γ/cc.	5 γ/cc.					
Normal	5	221	233	283	259	1.2 ± 0.06	285	595	2.1 ± 0.11
Estrogen	5	164	194	209	202	1.3 ± 0.05	224	518	2.3 ± 0.11
Gonadotropin	5	261	275	295	283	1.1 ± 0.03	580	636	2.0 ± 0.12

"P" value of difference between the effect of the enzyme on the spread of ink in estrogen-treated and gonadotropin = 0.002.

"P" value of difference between the effect of the enzyme on the spread of ink in normal and estrogen-treated = 0.10 (not significant).

"P" value of difference between the effect of the enzyme on the spread of hemoglobin in estrogen and gonadotropin-treated = 0.04.

"P" value of difference between the effect of the enzyme on the spread of hemoglobin in normal and estrogen treated = 0.11 (not significant).

between the skin of estrogen-treated and normal rabbits. This greater hydrolysis of hyaluronic acid by its enzyme in animals under the influence of estrogen than in rabbits under the effects of gonadotropin may be due to a number of variables. The interstitial pressure at the site of injection of the hyaluronidase, the concentration of the substrate, and its resistance to hydrolysis by the enzyme or the modification of the latter by the respective hormones, may be concerned.

A further attempt to differentiate the hyaluronic acid content of the skin of the above three groups of rabbits by staining the skin for metachromasia, as described by Wislocki and Dempsey (17), yielded no significant observations.

THE EFFECT OF ESTROGEN AND CHORIONIC GONADOTROPIN ON TISSUE AND VASCULAR PERMEABILITY

It is established that in the fowl estrogen causes a marked increase in the calcium level of the blood. In the present studies there was a slight increment of

the average concentration of the total serum calcium in estrogen-treated rabbits. Conversely, in rabbits under the influence of gonadotropin these calcium levels were somewhat lower than in control litter mates. However, these effects were not statistically significant. Nevertheless, bearing in mind the work of Chambers and Zweifach (18), which demonstrated that calcium reduces capillary permeability, an attempt was made to determine whether estrogen and chorionic gonadotropin affect this property. The capillary permeability was assayed in an indirect manner, using the method of Menkin (19).

The same group of 15 rabbits used in the above hyaluronidase experiment was tested two days later in the following manner. Three tenths cc. of the supernatant fluid of a centrifuged pleural exudate of one day's duration, derived from a dog treated with turpentine, was injected intracutaneously into each of the 15

TABLE 5

The effect of estrogen and chorionic gonadotropin on tissue and vascular permeability

Rabbit number	Normal: Tissue permeability: Spread of intracutaneous irritant in: 15 minutes	Normal: Tissue permeability: Spread of intracutaneous irritant in: 1 hour	Normal: Vascular permeability; intensity of color	Estrogen-treated: Rabbit number	Estrogen-treated: Tissue permeability: Spread of intracutaneous irritant in: 15 minutes	Estrogen-treated: Tissue permeability: Spread of intracutaneous irritant in: 1 hour	Estrogen-treated: Vascular permeability; intensity of color	Gonadotropin-treated: Rabbit number	Gonadotropin-treated: Tissue permeability: Spread of intracutaneous irritant in: 15 minutes	Gonadotropin-treated: Tissue permeability: Spread of intracutaneous irritant in: 1 hour	Gonadotropin-treated: Vascular permeability; intensity of color
A10=29	182	242	+++	A10=9	171	277	++	A10=11	292	338	+++
A10=42	231	337	+++	A10=31	158	249	++	A10=32	184	275	++++
A10=69	297	404	+++	A10=66	194	226	++	A10=67	219	413	++++
A10=82	225	329	++++	A10=80	151	206	+++	A10=81	147	250	++
A10=71	187	253	++	A10=72	154	212	+++	A10=72	500	626	+++++
Average...	225	313	+++	Average	166	234	+++±	Average	269	381	++++±

rabbits in two sites over the chest wall. Immediately thereafter, 15 cc. of a one per cent solution of trypan blue in saline were injected intravenously. Within a few minutes the site of injection of exudate became much more deeply colored than the rest of the skin. This was evidently a consequence of extravasation of dye into the area of increased vascular permeability penetrated by the leukotaxine containing irritant (19). These observations are presented in table 5, where the average of the two areas of extravasated dye in each rabbit is recorded fifteen minutes and one hour after the intravenous injection of the trypan blue. The intensity of the color of the dye at the site of extravasation is recorded as observed two hours after its intravenous administration.

It is apparent that in the estrogen-treated rabbits the exudate spreads over a smaller area than in the control litter mates, whereas in the rabbits under the influence of gonadotropin the irritant spreads over a greater area. This can be explained most readily as the result of the reduced permeability of the connective

tissue in estrogen-treated rabbits on the one hand, and of the increased permeability of this tissue in gonadotropin-treated rabbits on the other.

The intensity of color of these areas of trypan blue extravasation which apparently mirrors the amount of dye exuding from the vessels into the tissues, tends to be less in the estrogen-treated rabbits and more in the gonadotropin-treated animals than in the corresponding litter mate controls. This interpretation is plausible, for this difference could not be accounted for by variations in the rate of removal of dye from the site of extravasation by the tissues, as estrogen would aid its accumulation and gonadotropin would increase its dispersion.

TABLE 6

The effect of estrogen on the circulating lymphocytes of normal and tuberculous inbred rabbit litter mates

GROUP	RABBIT NUMBER	ABSOLUTE NUMBER OF LYMPHOCYTES PER MM³*			
		Before infection		After infection	
		Prior to estrogen treatment of group 2	20 days after estrogen treatment of group 2	11 days after infection	103 days after infection
(1) Not treated with estrogen	C8=2	4,392	4,520	4,579	4,704
	C8=17	4,858	4,185	3,750	5,088
	C8=19	4,772	3,738	5,481	7,627
	C8=23	5,236	3,953	2,525	4,630
	Average...	4,690	4,049	4,084	5,512
(2) Treated with estrogen	C8=1	4,720	2,150	2,208	4,802
	C8=16	4,982	4,683	1,515	1,340
	C8=18	4,862	2,247	1,980	3,032
	C8=24	2,215	2,072	972	2,480
	Average...	4,197	2,788	1,669	2,914

* Per cubic millimeter of free flowing, venous blood.

Thus additional evidence has been given for the view that the permeability of the connective tissue of estrogen-treated rabbits is reduced, whereas that of litter mates under the influence of gonadotropin is increased. Furthermore, there is suggestive indirect evidence that the permeability of the blood vessels may be similarly affected in opposite directions by these two hormones.

THE EFFECT OF ESTROGEN ON THE ADRENALS

During the course of these studies it was found that in estrogen-treated rabbits the absolute number of circulating lymphocytes was usually markedly reduced. This is illustrated in table 6. It will be seen that both in normal and tuberculous rabbits under the influence of the hormone the lymphocytes are conspicuously diminished as compared with those in normal or tuberculous litter mates which were not treated with estrogen.

In view of the demonstration by White and Dougherty (4) that the oxygenated corticosterones, mobilized by the adrenocorticotrophic hormones of the pituitary, cause the dissolution of lymphocytes, it was thought possible that the reduction of circulating lymphocytes in animals under the influence of estrogen may be due to an indirect stimulation of the adrenal cortex by this hormone. Hence studies were undertaken to determine the effect of estrogen on the adrenals.

Twenty-two mice were divided into two groups. Ten animals received three weekly subcutaneous injections of 5 γ of α-estradiol dipropionate in 0.1 cc. of sesame oil. This dose of hormone is equivalent on a body weight basis to that received by the rabbits in which the progress of tuberculosis was markedly retarded by this treatment. Twelve control mice received the same amount of sesame oil without the hormone. Two to four days after the last injection, all the animals were killed, and their total body weight and the weights of their adrenals, thymus, and uterus were determined. Sagittal frozen sections of the adrenals were stained with Sudan III and the width of the cortex was measured in arbitrary units by an ocular micrometer.

To determine the effect of estrogen on the adrenals of rabbits, 5 litter mate pairs, all past the age of eight months, were divided into two groups. One received three weekly subcutaneous injections of 0.5 mg. of α-estradiol diproprionate subcutaneously in 0.5 cc. of sesame oil. The remaining 5 litter mates received the same weekly amounts of oil without the hormoue. Another group of 4 litter mate pairs, seven months or older, were similarly divided and treated with the same amount of estrogen or oil, respectively, at the same intervals, for eight weeks. The first group was killed ten days after the last injection of estrogen. The second group was killed seven days after the last injection of the hormone. As the effect of three or eight weekly injections of estrogen on the adrenal weight was not significantly different, the two groups are treated as one.

To determine the effect of prolonged estrogen treatment on the adrenals of tuberculous rabbits, the weight of these organs at death was measured in 11 litter mate pairs, all simultaneously infected with the same suspension of virulent bovine tubercle bacilli. Each of the pair had been treated with estrogen or sesame oil, respectively, for fifteen weeks or more. The results obtained in these three experiments are summarized in table 7. The average weight of the adrenals is recorded in milligrams per 100 grams of body weight together with the standard deviation of the mean and the "P" value according to the "Student" statistical evaluation for each of the items compared.

It will be seen that the weight of the adrenals of mice given three weekly injections of estrogen is significantly increased. This weight enhancement is due to an increment in the width of the cortex which was 2.2 ± 0.5 micrometer units in the controls, and 2.9 ± 0.19 in the estrogen-treated mice. The difference was statistically significant with a "P" value of 0.002. This confirms the observation of Selye (3). It is noteworthy, as Selye had found, that estrogen causes a marked atrophy of the thymus. The weight of this organ in normal mice was 177 ± 22 mg. per 100 grams of body weight, while that of the estrogen-treated animals was 58 ± 5, with a "P" value of 1.000. Thus, mice given the amount

of estrogen effective in retarding the progress of tuberculosis in rabbits undergo the "alarm reaction" and respond with the general adaptation syndrome as described by Selye (3).

However, rabbits given the same amount of estrogen on a body weight basis for three to eight weekly injections, showed a slight but statistically insignificant increment in their adrenal weights as compared with litter mates treated with the same amount of oil without the hormone. On the other hand, prolonged treatment with this amount of estrogen in tuberculous rabbits is associated with a relative diminution of the adrenal weights as compared with the values for tuberculous litter mates not under the influence of the hormone. This reduction in the adrenal weights is of marginal statistical significance for the "P" value is 0.024. Thus, prolonged treatment with estrogens tends to diminish the size of the adrenals. The meaning of this observation has not been elucidated.

TABLE 7

The effect of estrogen on the weight of the adrenals of normal and tuberculous animals after varying intervals of treatment

NUMBER OF WEEKLY INJECTIONS	ANIMALS USED	NUMBER OF ANIMALS AND THEIR AVERAGE WEIGHT IN GRAMS				WEIGHT OF ADRENALS IN MG. PER 100 GRAMS OF BODY WEIGHT		
		Control		Treated		Control	Treated	"P" value of difference
		Number	Weight	Number	Weight			
3	Mice	12	18.9	10	19.1	38 ± 2.3*	51 ± 3.8†	0.004
3 to 8	Normal rabbits	9	3,390	9	3,170	10.8 ± 1.0	12.5 ± 1.4	0.166
15 or more	Tuberculous rabbits	11	2,320	11	2,630	23.4 ± 2.5	17.1 ± 1.7	0.024

* Width of cortex 2.2 ± 0.05.

† Width of cortex 2.9 ± 0.19. "P" value of difference between the width of the cortex of the adrenals in normal and estrogen-treated mice = 0.002.

There is, however, one striking observation recorded in table 7. The adrenal weight of untreated tuberculous rabbits is more than twice that of normal litter mates, with a "P" value of 1.000. This increase in milligrams of adrenal weight per 100 grams of body weight cannot be ascribed to the loss of body weight due to the tuberculosis and a failure of corresponding loss in adrenal weight. The average loss of weight of the tuberculous rabbits was only 31 per cent, whereas the gain of the adrenals constituted 117 per cent. Clearly, therefore, tuberculosis as such causes an increase in the size of the adrenal and the infection induces an hypertrophy in the adrenal cortex, for the medulla is not affected by the disease. This would suggest that the cortex of the adrenals exercises an important function in the course of tuberculosis. A similar, though statistically less significant, increase in the adrenal weights of estrogen-treated tuberculous rabbits as compared with hormone-treated nontuberculous animals was noted.

Not only is there no constant weight increase of the adrenals in response to estrogen but frozen sections stained with Sudan III of these adrenals, as well as of those of litter mates under the influence of gonadotropin, revealed no consistent differences in the distribution or staining intensity of the lipoids in the several cortical zones of these three groups of animals. Thus, no anatomical evidence of altered function of the adrenal cortex was found on administration of estrogen or gonadotropin.

As adrenal hypertrophy does not regularly follow estrogen treatment in rabbits, the observed reduction in circulating lymphocytes in these animals could not be attributed to an enhanced adrenocortical function. Sayers (20) demonstrated, however, that the corticotrophic hormone of the pituitary markedly reduces the ascorbic acid content of the adrenals, and that the concentration of this substance in the adrenal is a reflection of the corticotrophic funtion. Furthermore, White and Dougherty have caused the dissolution of lymphocytes with corticotrophic

TABLE 8

The concentration of ascorbic acid in the adrenals of normal, estrogen-treated, and gonadotropin-treated litter mates of the inbred rabbit family A

UNTREATED		ESTROGEN-TREATED		GONADOTROPIN-TREATED	
Rabbit number	Mg/100 mg. of adrenal	Rabbit number	Mg/100 mg. of adrenal	Rabbit number	Mg/100 mg. of adrenal
1	0.094	1	0.075	1	0.089
2	0.113	2	0.091	2	0.092
3	0.124	3	0.107	3	0.098
4	0.133	4	0.120	4	0.106
5	0.152	—	—	—	—
Average.....	0.123 ± 0.019	Average	0.098 ± 0.017	Average	0.096 ± 0.006

"P" value of difference between normal and estrogen-treated = 0.04.
"P" value of difference between normal and gonadotropin-treated = 0.01.

hormone in animals with intact adrenals. Hence it was thought that the concentration of ascorbic acid in the adrenals of control and estrogen-treated rabbits might throw some light on the behaviour of the lymphocytes in these animals.

Accordingly the concentration of reduced ascorbic acid in the adrenals of normal estrogen-treated and gonadotropin-treated litter mates was determined by the method of Rowe and Kuether (21). The results are presented in table 8.

It is evident that the ascorbic acid content of the adrenals of both estrogen and gonadotropin-treated animals is usually less than that of normal litter mates. This would suggest that the adrenals of animals under the influence of either hormone are subjected to increased corticotrophic stimulation. While the data are within the range of statistical significance, they are nevertheless too scanty for definite conclusions.

Further evidence for altered adrenocortical function in rabbits under the influence of these hormones was sought in the glycogen deposition of their livers as this process is a function of the adrenal oxygenated corticocosterones.

For this purpose the livers of the same group of 15 rabbits used in the above experiment on hyaluronidase were stained for glycogen by the Bauer-Feulgen method (22). It will be remembered that these rabbits consisted of three groups of litter mates. Five of these were under the influence of estrogens, 5 had been treated with chorionic gonadotropin, and 5 served as controls. All these rabbits had been on a constant diet for a long time and were killed on the same day. The first group of rabbits had received their third weekly injection of estrogen ten days before death; the second group had received their third intravenous dose of gonadotropin five days before they were killed. The rabbits in these three groups were of uniform weight and nutrition at the time of death, the average weight ranging between 3.1 to 3.4 kg. The estimate of glycogen deposition in each of these rabbits was based on the extent and intensity of the staining reaction.

While there was considerable variation in the deposition of glycogen in the individual rabbits of each of the three groups, it was nevertheless evident that the least glycogen deposits were found in the rabbits under the influence of gonadotropin. In some rabbits under the influence of estrogen the most extensive deposits of glycogen were found, though the overall difference between these rabbits and their litter mate untreated controls was not significant.

Thus no clear evidence has been obtained to demonstrate that estrogen increased the adrenocortical function in the rabbit. It must be stated, however, that in tuberculous rabbits under the influence of estrogen the adrenal cortex always appeared intact. In the adrenal cortex of tuberculous litter mates examined at the same interval after the same inoculation, but not treated with estrogen, there were often areas of necrosis, hemorrhage, and leukocytic infiltration, as may be seen in figures 1 and 2, Plate I.

That the administration of estrogen may affect other glands of internal secretion is suggested by the observation that, in more than half of the tuberculous rabbits under the influence of the hormone, the epithelium of the acini of the thyroid gland was definitely more cuboidal and the colloid accumulation less pronounced than in the untreated tuberculous litter mates.

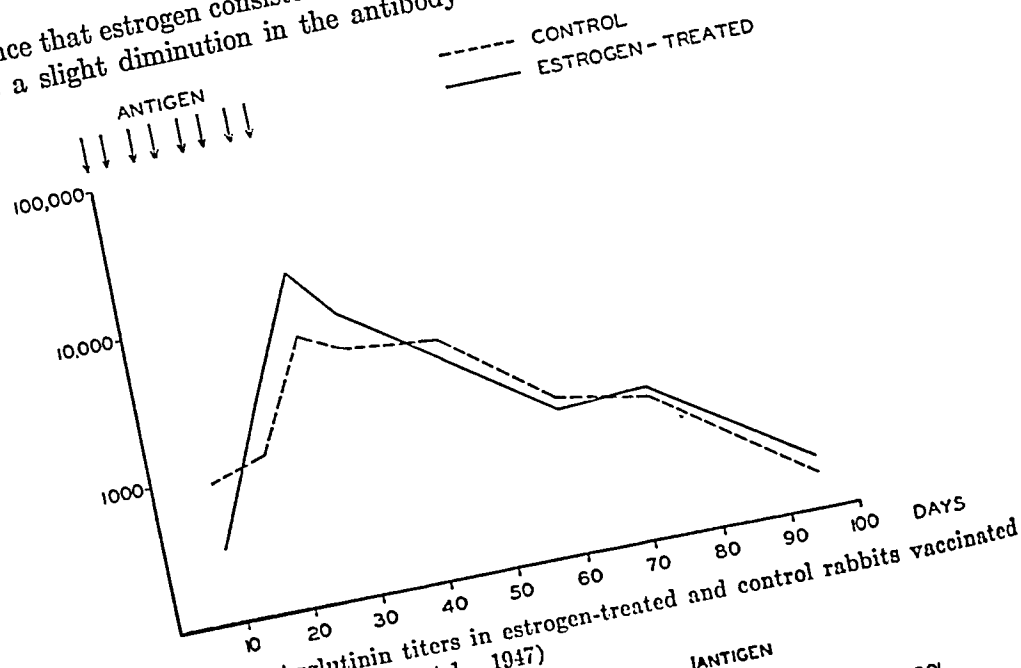
ESTROGEN AND ANTIBODY FORMATION

As stated above, estrogen causes a marked depression of circulating lymphocytes. There is considerable evidence that lymphocytes play a role in antibody formation. It has also been claimed that the administration of oxygenated corticosterones, which cause the dissolution of lymphocytes, enhances antibody formation (23). Moreover, von Haam has reported that theelin increases agglutinin production against the pneumococcus (24). Consequently an attempt was made to determine whether estrogen-treated rabbits produce antibody more rapidly and to a higher titer than normal litter mates.

Three such trials were made, two with chloroform-killed typhoid bacilli as antigen and one with alcohol-killed dysentery bacilli. The results are summarized in figures 1, 2 and 3. Each point plotted in these three figures represents the geometric mean of the antibody titer of 5 rabbits.

It is clear that, irrespective of the number of antigenic stimulations, there is no

evidence that estrogen consistently affects antibody production. There appears to be a slight diminution in the antibody titer of rabbits under the influence



TEXT FIGURE 1. Agglutinin titers in estrogen-treated and control rabbits vaccinated with typhoid bacilli. (Experiment 1. 1947)

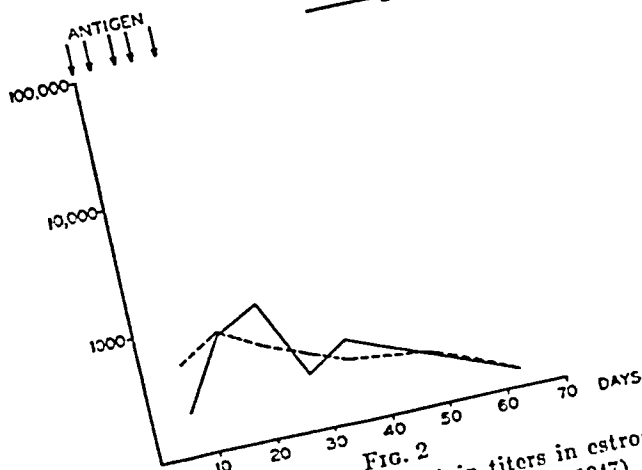


FIG. 2

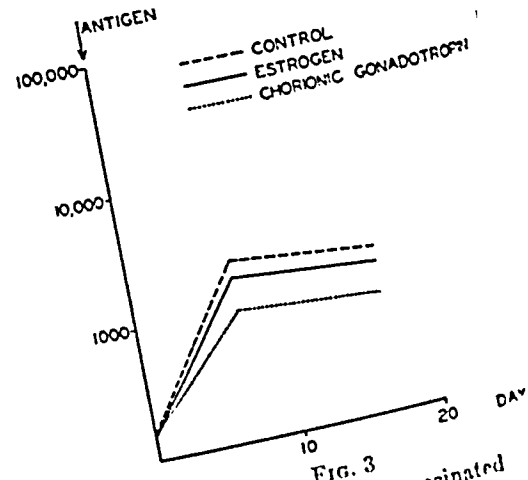


FIG. 3

TEXT FIGURE 2. Agglutinin titers in estrogen-treated and control rabbits vaccinated with typhoid bacilli. (Experiment 2. 1947)

TEXT FIGURE 3. Agglutinin titers in estrogen-treated, chorionic gonadotropin-treated, and control rabbits vaccinated with dysentery bacilli. (Experiment 3. 1948)

of gonadotropin. The difference, however, is not sufficiently constant or pronounced to be significant.

These observations indicate that differences in antibody formation in animals

under the influence of these two hormones do not account for their respective effects on the tuberculous process.

THE EFFECT OF ESTROGEN ON AMYLOID DEGENERATION

As is well known, chronic tuberculosis in man may be associated with extensive amyloid degeneration. Likewise, in rabbits dying from a slowly progressive tuberculosis, there is a severe amyloid degeneration of the malpighian corpuscles of the spleen. Thus the finding that tuberculous rabbits under the influence of

TABLE 9

The effect of estrogen and chorionic gonadotropin on the development of amyloid degeneration and its correlation with the extent of the existing tuberculosis

GROUP	RABBIT NUMBER	EXTENT OF AMYLOID DEGENERATION	EXTENT OF TUBERCULOSIS	GROUP	RABBIT NUMBER	EXTENT OF AMYLOID DEGENERATION	EXTENT OF TUBERCULOSIS
Untreated controls	C6-54	+++	10.0	Estrogen treated	C6-52	0	3.0
	C7-10	+++	20.0		C7-9	0	15.0
	C7-27	+++	17.5		C7-26	+	9.5
	C7-8	+	15.0		C7-7	0	9.0
	C7-29	++++	15.0		C7-28	0	6.5
	C7-13	+++	19.5		C7-12	0,TB*	8.5
	C7-16	+++	13.5		C7-15	±	8.0
	C7-36	+++	27.0		C7-40	+±	16.5
	A10=30	+++	22.0		A10=27	trace	14.0
	A10=23	0	10.5		A10=21	+	19.5
	A10=19	+++	24.5		A10=16	0,TB*	9.0
	A10=26	++	16.5		A10=25	+±	17.5
Untreated controls	A7=29	+++	12.0	Gonadotropin treated	A7=24	+++	17.5
	A8=22	++	16.0		A8=20	+++	16.0
	A8=38	++++	7.0		A7=39	+++	16.5
	A8=45	++++	7.5		A9=56	++	22.0
	A9=54	0	8.0		A9=63	0	18.0
	A9=61	0	13.0		A9=43	+++	9.0
	A9=40	++++	13.0		A9=87	+++	16.0
	A9=44	0	1.5				
	A9=57	0	5.5				

* Tuberculosis present.

estrogen either failed to show any amyloid degeneration at all, or were affected by it only to a very slight degree, appeared to be of considerable significance. Moreover, in untreated litter mates simultaneously infected with the same suspension of virulent bovine tubercle bacilli by the same route, which died or were killed at the same time, the degeneration was usually extensive and frequently had practically replaced most of the splenic parenchyma. This is illustrated in figures 3 and 4 of Plate I.

That this effect is specific for estrogen is indicated in table 9, which records the extent of amyloid degeneration in estrogen-treated and chorionic gonadotropin-

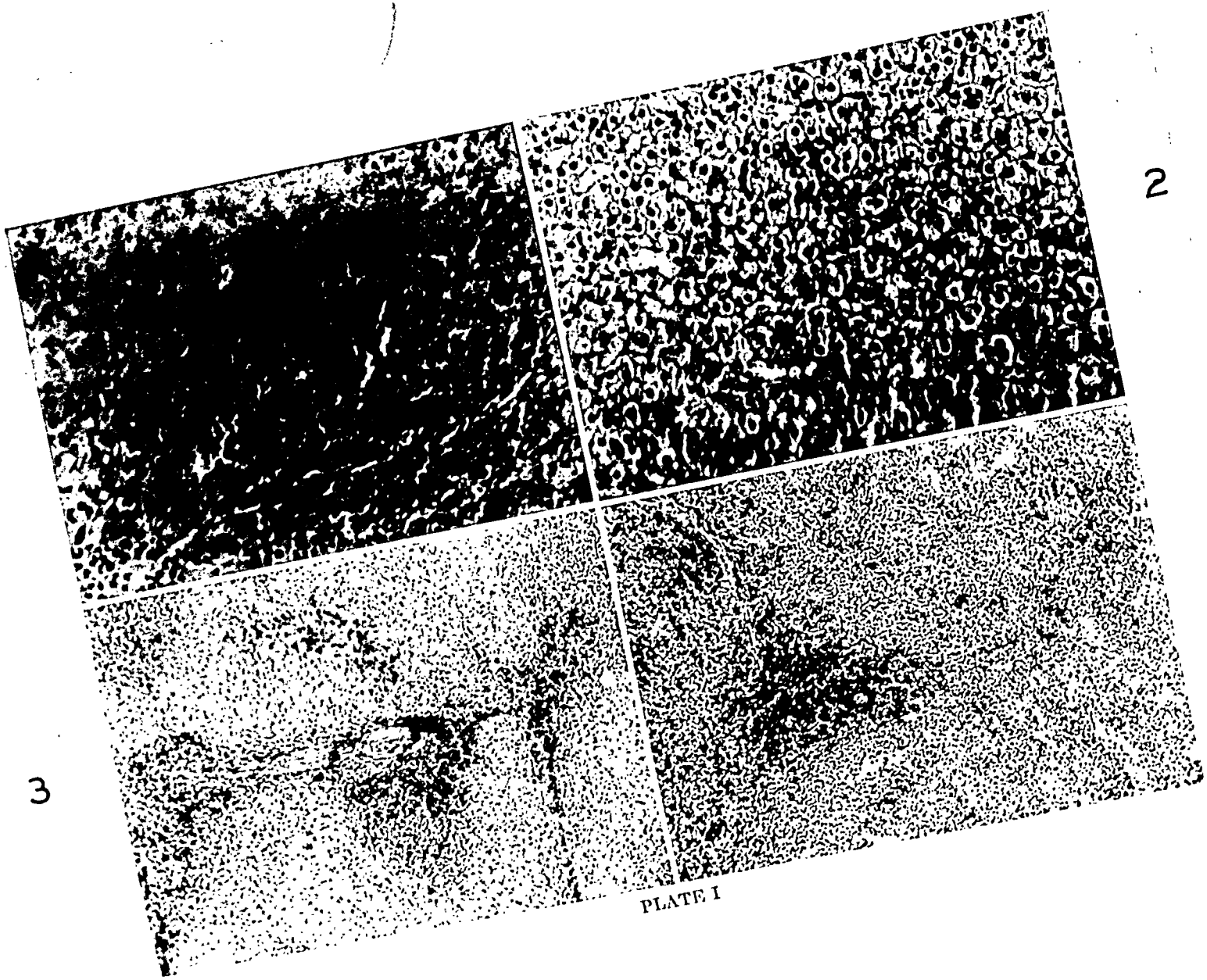


PLATE I

treated tuberculous rabbits, on the one hand, and in their untreated litter mate tuberculous controls, on the other. It will be seen that chorionic gonadotropin far from reducing the extent of amyloid degeneration actually tends to enhance it, whereas estrogen markedly suppresses this tissue alteration. As estrogen tends to retard the tuberculous process and gonadotropin tends to enhance it, it may be claimed that the amyloid degeneration is a result of the extent of the tuberculosis, and that estrogen by suppressing the tuberculosis also suppresses this degeneration. For this reason table 9 includes an estimate of the extent of the tuberculosis in each of these rabbits. This is assigned a numerical value as follows: the severity of the disease in the lung, pleura, and kidney is given a single number which is the sum of the degree of "plus" involvement in each, one plus being assigned the value of one. This sum also includes the number of other organs metastasized. One digit is given to each organ invaded. An examination of this table will reveal that, while there is a certain correspondence between the extent of the tuberculosis and the severity of the amyloid degeneration, the reduction in this degeneration of estrogen-treated rabbits is entirely out of proportion to the diminution of the tuberculosis in these animals.

Discussion

It has been shown in these studies that the administration of large amounts of estrogen to sexually mature highly inbred rabbits retarded the progress of tuberculosis at the site of inoculation in the skin and markedly diminished its dissemination to the internal organs as compared with that in untreated litter mates. In contrast, the induction of successive crops of corpora lutea in the ovary during the early phase of the infection by the periodic injection of chorionic gonadotropin, enhanced the disease at the portal of entry in the skin and increased its spread to the viscera. It has been shown in a previous study (25) that the chief difference in the pathogenesis of the disease in genetically resistant and susceptible rabbits is the degree of localization of the infection. Resistant animals restrict the disease to the portal of entry; susceptible rabbits permit it to spread. Therefore, the present investigation shows that, by altering the internal environment of the tissues of rabbits of the same hereditary resistance to tuberculosis by means of intense exposure to different naturally occurring hormones, one of the fundamental variants in the pathogenesis of the disease in rabbits of different natural resistance has been simulated. However, while there is evidence that

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PLATE I

Magnification of figures 1 and 2 approximately × 440, and of figures 3 and 4 approximately × 100

FIG. 1. (Upper left) Adrenal of control rabbit C7-29 at death. There is a large area of necrosis in the cortex.

FIG. 2. (Upper right) Adrenal cortex of estrogen-treated litter mate C7-28.

FIG. 3. (Lower left) Spleen of control rabbit C7-13. The pulp is largely replaced by amyloid degeneration.

FIG. 4. (Lower right) Spleen of estrogen-treated litter mate C7-12. There is no amyloid degeneration.

in naturally resistant animals the mononuclear phagocytes acquire the power to destroy tubercle bacilli more rapidly and effectively than the cells of susceptible animals, estrogen does not affect the growth and destruction of the bacilli in the tissues.

Since it was found in the study cited (25) that the most resistant family acquired allergic sensitivity on stimulation with heat-killed tubercle bacilli more rapidly and intensely than the most susceptible family, the effect of estrogen and chorionic gonadotropin on tuberculin sensitivity was studied. It was found that estrogen markedly suppressed the inflammatory response of the skin to tuberculin in rabbits which had been sensitized by active tuberculosis or by treatment with heat-killed tubercle bacilli. However, this seemingly depressing effect of estrogen on allergy was confined to the skin. The extent of caseation in the internal organs, the leukocytic response to the subcutaneous injection of tuberculin, and the sensitivity of the cells to tuberculin in tissue culture were not diminished by the hormone. Furthermore, even the lowered skin sensitivity to tuberculin could be restored by withdrawal of the hormone. The underlying allergizing mechanism was not affected by estrogen. The hormone merely masked an existing undiminished allergic skin sensitivity. Estrogen reduces the inflammatory response of the skin to bacterial toxic agents in general and, among these, to tuberculin, which is toxic to sensitized animals. Furthermore, in experiments not detailed it was found that chorionic gonadotropin did not affect the rate or intensity of development of allergic irritability of inbred rabbits treated with heat-killed tubercle bacilli.

Thus the mechanism whereby estrogen tends to localize the infection and gonadotropin tends to spread it differs from that by which naturally resistant rabbits restrict the infection to the portal of entry and susceptible animals permit its dissemination.

It was noted above that ovariectomy, which has no effect on the tuberculous process, and gonadotropin, which enhances the disease, both are associated with a lower skin sensitivity to tuberculin during the course of the infection. Therefore, the lower inflammatory irritability of the skin of estrogen-treated rabbits to tubercle bacilli and their products does not account for the retarding effect of the hormone on the disease.

It has been shown in these and previous studies that the spread of particles in the connective tissue is reduced by estrogen and increased by gonadotropin. In order to determine the mechanism whereby estrogen reduces the permeability of the connective tissue and thus hinders the dissemination of the disease from the portal of entry, the water content of the skin was investigated. It was found that the total concentration of water in estrogen-treated rabbits was not increased. However, the distribution of the water between the extra- and intracellular compartments was not assayed. Taylor and Sprunt determined the extracellular fluid of the skin of rabbits under the influence of this hormone by the thiocyanate method (26) and found a significant increase. This would suggest that, as a result of the swelling of the connective tissue elements caused by estrogen, their turgescence is increased and their permeability to particles reduced.

That the tautness of the connective tissue may be an important factor in its permeability was suggested by the following observation. Hyaluronidase enhanced the spread of particles to a greater extent in the skin of estrogen-treated rabbits than in that of litter mates under the influence of gonadotropin. As stated in the body of this paper, this may be due to a number of variables but the simplest explanation is afforded by the work of Hechter (16). He demonstrated that the spreading effect of hyaluronidase is determined not only by its concentration but also by the volume and pressure of the intradermal injection. The greater the interstitial pressure at the point of introduction of the enzyme, the more widely will the enzyme spread in the tissues, and the greater will be the extent of the hydrolysis of the hyaluronic acid matrix of the skin. As estrogen reduces permeability and gonadotropin enhances it, it is clear that a given volume of fluid injected into the skin of an estrogen-treated rabbit will be under a greater pressure than in an animal under the influence of the gonadotropin. Therefore, the hyaluronidase will diffuse to a greater degree in the estrogen-treated than in gonadotropin-treated animals. On this basis, no assumption need be made as to the effect of these hormones on the enzyme or its substrate to explain the observations. Therefore it may be inferred that one factor in the permeability of the connective tissue is the turgidity of its elements.

Finally, by the use of Menkin's technique presumptive evidence was obtained which suggests that the vascular permeability of the skin may be reduced by estrogen and increased by gonadotropin. All these observations are in harmony with the observed restricting effect of estrogen on the dissemination of the disease from the portal of entry in the skin and its enhancement by gonadotropin.

Physiologic doses of combined progesterone and estradiol, which also enhance the spread of particulate matter in the skin, do not increase the dissemination of tuberculosis in the body. This would suggest that the action of corpora lutea engendered by gonadotropin was not completely simulated by the administered hormones. On the other hand, the failure of estrogen to restrict the tuberculosis in sexually immature rabbits regularly would imply that other changes besides reduced connective tissue permeability may play a role in this hormone's capacity to retard the disease.

It has been shown by Selye that folliculoids induce the adaptation syndrome. In mice, the administration of estrogen in amounts comparable to that used in rabbits induced hypertrophy of the adrenal cortex and atrophy of the thymus. In rabbits there is a reduction of circulating lymphocytes but no increase in adrenal weights as a result of estrogen. Moreover, no definitive evidence was obtained by the study of liver glycogen deposits of increased adrenocortical function in the rabbit as a result of estrogen treatment. Lipoid stains of the adrenals of these animals also failed to indicate any alteration of cortical function.

While the adrenals of rabbits show no hypertrophy in response to estrogen treatment, they nevertheless appear to contain less ascorbic acid than the adrenals of untreated litter mates. This fact suggests that the glands are subjected to increased corticotrophic stimulation of the pituitary. This acid was also reduced, however, in animals under the influence of gonadotropin. Hence the opposite effects of these two hormones on the tuberculous process cannot be

ascribed to their adrenocortical influence on this basis. The increased secretion of ascorbic acid by the adrenals may be a factor in the reduction of the tuberculin sensitivity of their skin, for Steinbach (27) showed that sensitized guinea pigs may be protected from tuberculin shock with ascorbic acid.

In view of recent claims that antibody production is enhanced by the administration of cortical hormones, it is noteworthy that neither estrogen nor gonadotropin significantly affected agglutinin formation against the typhoid or dysentery bacillus. While there is no clear evidence that these sex hormones affect the disease via the adrenals, it must be emphasized that tuberculosis as such causes a marked hypertrophy of the gland, suggesting that it plays a significant role in the tuberculous process. In this relation it may also be stated that in one-half of the rabbits under the influence of estrogen there was some hyperplasia of the thyroid. This indicates that the administration of estrogen is not without effect on other endocrines.

One of the most impressive differences between estrogen-treated and control tuberculous rabbits was the marked reduction in amyloid degeneration in the spleen which is characteristic of rabbits dying from chronic tuberculosis. This could not be accounted for by the less extensive disease in the hormone-treated animals. Frequently the difference in the extent of the disease was only moderate in the two litter mates, whereas the amyloid degeneration was extensive in the control animal and completely absent in the estrogen-treated rabbit. This effect was specific for estrogen, as ovariectomy or progesterone and gonadotropin treatment had no effect on the degree of amyloid degeneration. The significance of this observation remains to be determined.

In the same category is the fact that, while prolonged estrogen treatment tends to reduce the adrenal weights, the incidence of degenerative processes in the adrenal, such as hemorrhage, leukocytic infiltrations, and focal necrosis is definitely reduced in estrogen-treated rabbits. What this means has not yet been ascertained.

Conclusions

1. Estrogen retards the progress of tuberculosis at the portal of entry in the skin and diminishes its dissemination to the internal organs in highly inbred rabbits chiefly by reducing the permeability of the connective tissue.

2. Chorionic gonadotropin enhances the disease at the portal of entry and its spread through the body mainly by increasing the permeability of the connective tissue.

3. Hyaluronidase exerts a greater spreading effect in estrogen-treated animals than in rabbits under the influence of gonadotropin.

4. Estrogen reduces the number of circulating lymphocytes of the blood. The intermediation of the adrenal cortex in this effect was not demonstrated.

5. Tuberculosis *per se* induces a marked hypertrophy of the adrenals in rabbits.

6. The reduction of the inflammatory responsiveness of the skin to the products of tubercle bacilli induced by estrogen is not a significant factor in this hormone's capacity to retard the tuberculous process.

7. Estrogen and gonadotropin exert no effect on antibody formation.

8. Estrogen markedly suppresses the development of amyloid degeneration in the spleens of tuberculous rabbits.

Sumario

Factores Constitucionales en la Resitencia a la Infección: III. Modo de Actuar del Estrógeno y de la Gonadotropina sobre la Evolución de la Tuberculosis

1. El estrógeno retarda el proceso tuberculoso en la vía de entrada en la piel y disminuye su diseminación a los órganos internos en los conejos muy entrecruzados, principalmente por reducir la permeabilidad del tejido conjuntivo.

2. La gonadotropina coriónica realza la enfermedad en la vía de entrada y su propagación por el cuerpo, principalmente por acrecentar la permeabilidad del tejido conjuntivo.

3. La hialuronidasa ejerce mayor efecto propagador en los animales tratados con estrógeno que en los conejos sometidos al influjo de la gonadotropina.

4. El estrógeno reduce el número de linfocitos en la circulación. No se demostró la intervención de la corteza suprarrenal en este efecto.

5. La tuberculosis evoca *per se* pronunciada hipertrofia de las suprarrenales en los conejos.

6. La disminución, evocada por el estrógeno, de la respuesta inflamatoria de la piel a los productos de los bacilos tuberculosos no constituye un factor significativo en la capacidad de dicha hormona para retardar el proceso tuberculoso.

7. El estrógeno y la gonadotropina no ejercen efecto alguno sobre la formación de anticuerpos.

8. El estrógeno suprime notablemente el desarrollo de degeneración amiloidea en los bazos de los conejos tuberculosos.

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LETTERS TO THE EDITORS

STREPTOMYCIN-DEPENDENT STRAINS OF MYCOBACTERIUM TUBERCULOSIS

To the Editors of the American Review of Tuberculosis:

Miller and Bohnhoff in 1947 (Science, 1947, *105* (2737), 620) were the first to demonstrate that bacterial variants dependent upon streptomycin for growth may appear in cultures otherwise sensitive to streptomycin. This observation has been confirmed since by a number of other investigators. Recently we have been successful in isolating several streptomycin-dependent variants of *M. tuberculosis*.

In a study of the chemotherapeutic value of various antituberculous agents, mice infected intracerebrally with the $H_{37}Rv$ strain of *M. tuberculosis* were cultured at the time of death on four media: (*1*) Dubos liquid medium, containing 0.02 per cent Tween 80 and 0.5 per cent albumin; (*2*) Dubos medium to which had been added streptomycin in a concentration equivalent to 500 γ of pure streptomycin base per ml.; (*3*) Herrold's egg yolk agar; and (*4*) Herrold's egg yolk agar to which streptomycin had been added in a concentration equivalent to 500 γ of pure base per ml. Incubation was carried out at 37° C for a period of six to eight weeks.

A total of 196 animals were cultured. Of these, 161 were positive for tubercle bacilli when cultured on the Dubos medium containing no streptomycin; 147 were positive when cultured on Herrold's egg yolk medium containing no streptomycin. On one or both of these media, 184 (95 per cent) of the 196 animals cultured were positive for tubercle bacilli. All of these strains were highly sensitive (<10 γ per ml.) to streptomycin in spite of the fact that many of the animals had received repeated injections of large dosages of streptomycin for thirty-one days following inoculation.

Of the 196 animals cultured, 13 were positive for tubercle bacilli when cultured in Dubos liquid medium containing 0.02 per cent Tween 80, 0.5 per cent albumin, and the equivalent of 500 γ of streptomycin base per ml. These same 13 animals all showed growth of streptomycin-sensitive tubercle bacilli when cultured in the absence of streptomycin. All of the strains isolated on the streptomycin-containing media at first grew only in the presence of high concentrations of streptomycin. On subsequent transfers *in vitro*, rapid and heavy growth appeared in all instances on media containing streptomycin, dihydrostreptomycin, mannosidostreptomycin, or streptomycin residues containing both streptomycin and mannosidostreptomycin. Faint growth occurred at this time, however, in all instances on streptomycin-free media, but subculture of these nondependent variants indicated that they could not be propagated repeatedly in the absence of streptomycin.

In all cases, the streptomycin-dependent strains of tubercle bacilli described above were highly resistant to streptomycin *in vitro*. The sensitivity of 8 of these strains to streptomycin ranged from 5,000 to 10,000 γ per ml., while 2 were sensitive to 2,500 γ per ml. The remaining 3 strains grew readily in concentrations of streptomycin equivalent to more than 500 γ of pure streptomycin base per ml. In all instances, the growth was less in the presence of low concentrations of drug, suggesting that the strains may well contain some cells growing more readily in the presence of higher concentrations of streptomycin.

In no instance were streptomycin-dependent variants isolated on Herrold's egg yolk medium containing streptomycin.

Of the 13 streptomycin-dependent strains isolated, 6 were tested for virulence and for their *in vivo* sensitivity to streptomycin. Mice belonging to the C57 or DBA strains were

infected intracerebrally with 0.01 cc. of a standardized culture of each strain. All cultures used for inoculation of mice were grown for ten days in the Dubos liquid medium containing 0.02 per cent Tween 80 and 0.5 per cent albumin. Cultures were diluted with sterile Dubos liquid medium immediately prior to use to a final density of 57 to 63 per cent transmission (Photovolt Lumetron No. 400). A portion of the infected mice were held as untreated controls while 10 received 4 mg. (total weight) of pure streptomycin sulfate daily by the subcutaneous route for a period of thirty-one days. All animals were observed for a period of six months. Animals were autopsied at the time of death and the presence of tuberculous lesions determined by gross pathological examination, culture for tubercle bacilli and by microscopic examination for acid-fast bacilli. The degree of pathogenicity was measured by the rate at which the mice succumbed to the tuberculous infection. Only one or 2 of the 6 strains showed a degree of virulence comparable with the original culture of $H_{37}Rv$ from which these streptomycin-dependent strains had been derived. The remaining strains possessed little virulence for mice, three to six months being necessary to produce death in a portion of the animals, while the remainder survived for a period of over six months. In no instance was virulence enhanced by the administration of streptomycin. Those strains possessing virulence, however, were sensitive to streptomycin *in vitro*.

At the time of death all animals showed gross evidence of tuberculosis. On culture from the lungs of a portion of these animals, both streptomycin-resistant and streptomycin-dependent strains of tubercle bacilli were isolated. In no instance were streptomycin-sensitive strains recovered. This is of interest in view of the fact that some of the cultures were recovered from streptomycin-treated animals in which the per cent survival was slightly greater than in the nontreated animals.

It is apparent from these observations that variants of *M. tuberculosis* dependent upon streptomycin for growth may appear in otherwise sensitive cultures. It is further apparent that dependency probably is associated closely with resistance in view of the fact that all dependent strains isolated were resistant to streptomycin *in vitro*, and on passage *in vitro* produced streptomycin-resistant and streptomycin-dependent strains only. The ability of microorganisms to adapt themselves in such manner as to permit their utilization of streptomycin as an essential factor for growth may contribute in part to the limited effectiveness of the drug as a chemotherapeutic agent.

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October 29, 1948.

TUBERCULOSTATIC ACTIVITY OF AUREOMYCIN IN VITRO AND IN VIVO

To the Editors of the American Review of Tuberculosis:

In view of the interest in the possibilities of aureomycin as a therapeutic agent in tuberculosis, it may be of value to present some preliminary results of experiments on the tuberculostatic effect of this drug *in vitro* and *in vivo*.

The growth inhibiting action of aureomycin[1] (Lederle Lot No. 7-8071 A, crystalline hydrochloride) on the laboratory strain H37 Rv was tested in four different types of fluid medium. After eleven days of incubation, growth was inhibited by 5 γ of aureomycin per cc. in Tween-albumin medium; 2.5 γ per cc. in Proskauer and Beck's medium; 5 γ per cc. in Youmans' modification of the Proskauer and Beck medium, and 40 γ per cc. in the latter medium plus 10 per cent beef serum. At the sixteen day observation period, the minimal inhibitory concentration was 10 γ per cc. in Tween-albumin and Proskauer and Beck's medium, 20 γ per cc. in Youman's modification of Proskauer and Beck's medium, and over 40 γ per cc. in the medium with serum.

Twenty guinea pigs (average weight 500 Gm.) were infected subcutaneously with 0.02 mg. of the H37 Rv microorganisms and one-half of the group were started on aureomycin treatment ten days later. The drug was given by intramuscular injection at twelve hour intervals. Initial dosage was 0.2 mg. per day, which was increased gradually to 1.6 mg. per day. It was felt that the animals could not tolerate much more than this amount for they exhibited a marked weight loss and appeared to be in worse condition than the untreated controls subsequent to the third week of therapy.

All survivors were killed 58 days after infection. At this time, 7 of the treated animals had died, but only 2 of the controls had succumbed. A comparison of the amount of tuberculous disease observed grossly at death or sacrifice revealed no difference between the group treated with aureomycin and the untreated control group.

It would appear from these observations that aureomycin, in the dosage used, had no deterrent effect on the course of the tuberculous disease in guinea pigs.

It is realized that the aureomycin dosage was relatively small, but it compares favorably with the intramuscular dosage in man reported in the literature (0.6 to 3 mg. per kg. per day, Bryer *et al.*, J. A. M. A., 1948, *138:* 117; and Wright *et al.*, J. A. M. A., 1948, *138:* 408). Moreover, the dose used in the present experiments was the maximum tolerated dosage for the guinea pigs by this route. The response of tuberculous infection of animals of other species which might be able to tolerate higher doses of aureomycin cannot be predicted. The intramuscular administration of aureomycin to guinea pigs, however, had no effect upon the course of their tuberculous infections.

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[1] Supplied by courtesy of the Antibiotics Study Section, National Institute of Health, United States Public Health Service, Washington, D. C.

THE

AMERICAN REVIEW OF TUBERCULOSIS ABSTRACTS

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Tuberculous and Meningococcal Meningitis.—A patient with meningococcal meningitis was being treated adequately. Although the cerebrospinal fluid became sterile, the patient's condition suddenly took an unfavorable turn. Evidence of a coexisting tuberculous infection then was found and the patient died. The diagnosis was confirmed by necropsy. In retrospect, the fact that only 54 per cent of the cells in the cerebrospinal fluid were neutrophiles is of interest. The reported case is about the thirty-fourth in the literature.—*Coexisting tuberculous and meningococcal meningitis, E. A. Riley, New England J. Med., September 29, 1948, 239: 886.*—(A. G. Cohen)

Tuberculous Pericarditis.—Tuberculosis is recognized as a cause of constrictive pericarditis. Pockets of fluid or of inspissated residue have been found in the pericardium but a frank abscess containing tubercle bacilli has not been reported. Pleural effusion is a frequent concommitant and may be part of a polyserositis. A case is reported which progressed through the usual course of pericardial effusion, pleural effusion, and venous obstruction. At operation a tuberculous abscess was found, pericardectomy was done, and streptomycin was instilled locally and given parenterally after operation. A good clinical result was obtained and no sinus developed.—*Constrictive pericarditis with tuberculous intrapericardial abscess treated with streptomycin, J. A. Moore & J. D. Murphy, Ann. Surg., May, 1948, 127: 685.*—(*W. H. Oatway, Jr.*)

Pathogenesis of Industrial Pulmonary Disease.—The discussion is limited to subacute and chronic pulmonary disease resulting from the inhalation of particulate matter of low solubility. The inhalation of irritating dusts results in certain changes in the behavior of the lung which in some cases favor the retention of fine particles in larger numbers than usual. For example, there is a reduction in the pulmonary volume and an increase in the rate accompanied by bronchiolar constriction which, if maintained, leads to stasis of secretions, patchy atelectasis, and emphysema. Bronchiolar constriction is accompanied by an increase in residual air. Reduction of the capillary flow in atelectatic areas creates conditions favorable to the accumulation of phagocytes which have engulfed the dust. These phagocytes may die *in situ*, autolyze, and liberate dust which, if toxic, stimulates local fibrosis. Thus atelectasis and emphysema resulting from bronchiolar constriction favor markedly the local deposition of dust. The majority of the particles are carried by lymphatics to the hilar nodes but some enter the capillaries and flow into the systemic circulation. This is a protective mechanism as regards particles that have settled in the alveoli and the overall drainage rate is an important factor. The upper portions of the lung are first and most severely affected, suggesting that the rate of drainage is closely related to lung movement. This is consistent also with the observation that continued activity sometimes delays the onset and lessens the disability in some progressive pulmonary diseases. At present the solubility of the

particles is thought to be the most important factor in the production of pneumoconiosis. Certain critical considerations must be evaluated, however, before accepting this theory. (*1*) The chemical attributes of inhaled material must greatly affect the character and degree of tissue reaction. (*2*) The differences in solubility between active and inactive dusts have been classified only empirically as yet. (*3*) Toxic materials and ions which dissolve from the surface must be shown to differ quantitatively and qualitiatively and their solubility must be related to the pathogenicity of the particles. This had been shown to be generally true of silica. The determination of solubility of silicious materials is difficult but it has been shown that the solubility of quartz dust is inversely related to the particle size. Other unsolved problems include the effects of critical concentration on the rate and the course of development of the disease. It follows that the development of chronic pulmonary disease is dependent upon (*1*) the chemical characteristics of the dust, (*2*) the solubility rate of particles within certain ranges and (*3*) exposure to an adequate concentration of dust over a sufficient period of time. The rate of development will depend upon the adequacy of the drainage system as determined by previous history with respect to earlier infections and exposure to the irritant. Tissue reactions are seen peribronchially, in the pleura, perivascular tissues, interstitial tissues, alveolar wall, and along the lymphatics. Where phagocytes have accumulated and died, focal proliferation in the perilymphatic sheaths with an accumulation of phagocytes impairs lymphatic drainage. The outstanding pathology may be in the lung and the disturbance of lung function may dominate the picture but with long-continued, severe exposure lesions develop in the abdominal viscera.—*Pathogenesis of industrial pulmonary disease, W. Meeks, Radiology, June, 1948, 50: 755.—G. F. Mitchell*

Pulmonary Changes in Beryllium Workers.—Workers are exposed to beryl dust at the mines during the various processes of extraction of beryllium from the ore, the production of its salts, oxides and alloys, during the manufacture of fluorescent lamps, neon tubes, radio tubes, and in research and pharmaceutical plants. In all of these situations there has been definite evidence of disease among the employees with symptomatology, roentgenographic pattern, and pathology so similar that beryllium or one of its compounds is suspected as the cause. The incidence and severity of the disease are proportionate to the degree of exposure and chemical irritation. Most patients recover within a few weeks or months following removal from exposure but recurrence may occur without re-exposure and the disease may last months or years. The onset may be as long as six years after the last exposure. The onset is insidious and characterized by cough with occasional dull, burning substernal pain. Dyspnea follows the cough; cyanosis, low blood pressure and clubbing of the fingers are frequent physical findings. Anorexia and weight loss are common, usually accompanied by increasing fatigue. Physical findings indicate an absence of infection although the pulse rate is rapid. The pathology is that of an atypical pneumonitis, pulmonary edema, and hemorrhage. The lesions in the so-called chronic pulmonary granulomatosis are seen grossly as small pinhead-sized, whitish nodules throughout the lungs. Lymph nodes occasionally are enlarged and may show areas of granular, grayish-white necrosis. The earliest lesions consist of clumps of hystiocytes and moderate edema within the alveoli. Later the nodules of histiocytes are surrounded by a zone of lymphocytes and occasional plasma cells. The end stage is marked by an area of hyalinization. Only subacute and chronic forms of the disease have been recognized but it is possible that there is an acute type also. The roentgenographic findings are dependent upon the pathology present. A diffuse ground glass density appears in both lungs one to three weeks after the onset of symptoms, followed by areas of soft infiltrations are expanded by distinct linear markings and lobar enlargement. The infiltrations disappear and small

nodules are then seen. These disappear in one to four months and are usually gone before the symptoms subside. Some residual fibrosis may be seen. Most of the cases followed by the authors, some since 1942, were of the chronic type. Much hilar lymph node enlargement was found in these cases and was associated with upward displacement of the short septum and with basal emphysema. With the increase in basal emphysema more cases having clubbing of the fingers and cyanosis may appear. Seven cases developed small pneumothoraces. A few cases have shown a clearing of the granular and nodular densities, but some residual linear fibrosis was present and clinically there was evidence of subnormal pulmonary function. The size of the nodules varied in individual cases; in a few the original "sand-like" pattern was retained or simulated by a fine nodulation. Roentgenograms during acute exacerbations showed what is believed to be edema. The patient usually recovered with bed-rest and oxygen therapy and the roentgenographic findings returned to the pattern seen before the attack. Those who succumbed usually had cor pulmonale. The degree of involvement as seen roentgenologically is not always an indication of the degree of disability. Differential diagnosis includes a consideration of pulmonary sarcoidosis, miliary tuberculosis and silicosis. No specific mode of treatment has been found. Spectrography has been the only reliable method for determining such small amounts as 5 γ or less. It can be used to detect beryllium or beryllium products in the urine and to analyze dust concentrations in the plants.—*Delayed chemical pneumonitis or diffuse granulomatosis of the lung due to beryllium, S. A. Wilson, Radiology, June, 1948, 50: 770.—(G. F. Mitchell)*

Beryllium Pneumonitis.—A laboratory. worker developed a chemical pneumonitis three years after exposure to zinc beryllium manganese silicate used in making fluorescent lamps. The period of exposure was one year. The chief symptoms were cough, dyspnea and extreme loss of weight. The roentgenogram four months after the onset showed fine punctate lesions or gray markings throughout both lung fields. A film at eight months showed the same findings with some evidence of a reticular pattern. At seventeen months, there was more reticulation with small scattered nodules about 2 mm. in diameter. At twenty-one months, there was more definite fine nodulation. At thirty months, there was considerable fibrosis. A liver biopsy showed a granulomatous lesion. Clinically, there was no improvement three years after the onset.—*Delayed pneumonitis in a beryllium worker, J. N. Agate, Lancet, October 2, 1948, 2: 530.—(A. G. Cohen)*

Anthraco-silicosis.—Anthraco-silicosis, commonly called "miner's asthma", is a chronic disease due to breathing air containing dust formed in the various processes of mining and preparing anthracite coal. It is characterized anatomically by generalized fibrotic changes throughout the lungs, the presence of excessive amounts of carbonaceous and siliceous material, a compensating emphysema, and often by cardiac changes in the later stages of the disease. A review of some observations made in coal miners with anthraco-silicosis over a period of 27 years is presented. One thousand cases were studied from the standpoint of the age and build of the worker, the working conditions, years of exposure, and race. No race was found to be particularly resistant. A man must be exposed to a siliceous dust from five to seven years before changes in the lungs can be noted in the roentgenogram. A thorough history and roentgenographic examination, including fluroscopy, is essential for the diagnosis. Stereoscopic films are indispensible in the first and second stages of the disease. Infection is more frequent in the third stage of the disease and nonspecific infections in the first and second stages respond quite well to ordinary treatment. Tuberculosis is not a common complication in the first two stages of the disease nor under the age of fifty. Total and permanent disability has never been observed from anthraco-silicosis alone unless the dis-

ease has advanced well into the third stage. Evidence so far seems to indicate that the disease does not progress to the third stage if the patient is completely removed from exposure to the dust. Since anthraco-silicosis is a progressive, incurable disease, the only way to counteract it is to remove the silica from the atmosphere in which the men work. The men cannot be removed without closing the industry. The best method for removing the siliceous dust is a system of water sprays used before and after drilling and blasting.—*Anthraco-silicosis, W. J. Corcoran, Radiology, June, 1948, 50: 751.*—(*G. F. Mitchell*)

Lung Changes from Alumina Abrasives.—A review of the case histories of men who have been employed in the manufacture of alumina abrasives shows that there are now 34 well established and 38 early cases of distinctive pulmonary disease associated with this industry. The classification of well established disease is based on clinical and radiological evidence. All patients with symptoms have changes of moderate to extreme degree demonstrable by roentgenography. Others may have few symptoms but in each the roentgenogram shows a widened mediastinum with considerable parenchymal shadowing; such persons usually progress to a more serious type of disease. The longest period of exposure reported was nineteen years, the shortest eighteen months. The men who change the furnace pots and instructors had greater exposure to the hazard than binmen and cranemen. Several men in various plants have worked as long as nineteen years continuously as furnace feeders and have shown no roentgenographic changes to suggest that they are suffering from the disease. Disease from working in alumina abrasives was first discovered during the recent war but there is sufficient evidence to show that the disease was present prior to that time. A review of previous films has uncovered additional cases which have been classified as showing early disease. These were not previously reported. Seven of 34 well established cases died within a relatively short time after they were first seen. Three cases have been diagnosed and the men removed from exposure too recently to evaluate the results. The remaining 24 cases are presented as showing extension of the disease in 4 different degrees with distinctive roentgenographic changes. There is little evidence to prove that acute infection plays any major part in the progression of the disease.—*Further observations of lung changes associated with the manufacture of alumina abrasives, C. G. Shaver, Radiology, June, 1948, 50: 760.*—(*G. F. Mitchell*)

Acute Pulmonary Actinomycosis.—A twenty-year-old woman was successfully treated for acute pulmonary actinomycosis complicated by actinomycosis of the right pleural cavity. The pneumonic process appeared to respond to intramuscular penicillin, but this did not prevent the formation of empyema. After actinomyces had been identified in the pleural pus, penicillin by intramuscular injection was continued, together with sulfonamide therapy ("Elkosin" by mouth) and the pus was drained through an intercostal incision. Improvement from this point on was prompt but there was recurrence of empyema which necessitated drainage through a higher space. Convalescence then proceeded uneventfully and the patient has remained without recurrence for one year. In all, 1,425,000 units of penicillin and 250 Gm. of "Elkosin" were given. Local treatment of the empyema space with penicillin was not carried out although a review of the literature shows that this is effective in more chronic cases.—*Heilung eines Falles von Lungenaktinomykose mit postpenumonischem aktinomykotischem Empyem durch Bülau-Drainage und Penicillin-Elkosin Medikation, R. Fink, Schweiz. med. Wchnschr., January 10, 1948, 78:19.*—(*M. Marcus*)

Q-Fever.—Up to 1947 Q-fever had not been identified in Switzerland. Undoubtedly cases of virus-type pneumonia might have been diagnosed as such in the past if proper methods of identification of the rickettsial disease had been available. The author reports cases

observed in 1947 which could be proved to be identical with Q-fever by complement fixation tests. He was able to identify the disease among a family group of 4 people all of whom became ill within three days with a characteristic febrile illness accompanied by moderate to marked prostration and extensive pathological findings in the lungs. Another epidemic of this disease was obsered in a village where 50 cases of Q-fever pneumonitis were seen within four weeks. One of these cases was fatal. Thirty cases were observed in a boarding school in Geneva in June, 1947 and sporadic cases have been observed in the spring and fall of 1947 in the canton of St. Gallen. The illness was characterized by fever, severe constitutional symptoms, headache, and profound prostration. Physical examination was often negative but roentgenograms usually showed impressive consolidations in one or more lobes. The course was usually benign with resolution between the fifth to twelfth day, but pleural effusions and orchitis were frequently seen. The etiological agent is Rickettsia burneti and diagnosis is usually made by rising titers of the specific complement fixation test. A titer of more than 1:28 is diagnostic. The vector is believed to be Dermacenter andersoni which is commonly found on cattle but ticks were not encountered in the cases reported in this paper. Treatment of the illness is supportive and symptomatic.—*Q-fever (Queenslandfieber) in der Schweiz, O. Gsell, Schweiz. med. Wchnschr., January 10, 1948, 78: 1.*—(*M. Marcus*)

Bronchial Adenoma.—Of 217 primary bronchial tumors proved histologically at the Lahey Clinic since 1930, 15 (6.9 per cent) were adenomas. They were located principally in the major bronchi. They were seen by the bronchoscopist in 12 of 14 cases examined. Grossly the tumor appears as a rounded, pink or reddish-purple mass, usually attached by a broad base. It bleeds profusely with manipulation. There are 2 histological types, carcinoid and mixed (cylindroma); the latter is considered by some to have malignant potentialities. Cases of adenoma which underwent malignant degeneration with or without metastases have been reported. Local infiltrations are frequent. Sixty per cent of the cases were in patients less than 40 years of age. Eight were in females. The duration of respiratory symptoms prior to diagnosis ranged from eight months to six years. Cough is the most frequent symptom; it usually becomes progressively worse and eventually is productive. Hemoptysis is an outstanding symptom. A wheeze was found in 3 cases. Dyspnea was present in 4 cases and was the result of atelectasis, obstructive emphysema or mediastinal shift. Varying degrees of chest discomfort were noted and pulmonary infections were frequent. Abnormal physical signs varied with the pulmonary changes. The treatment is either local removal or pulmonary resection. Local removal is adequate provided that there is a pedicle and no extrabronchial extension, irreversible pulmonary damage or involvement of the carina.—*Bronchial adenoma, C. R. Souders & J. W. Kinasley, Jr., New England J. Med., September 23, 1948, 239: 459.*—(*A. G. Cohen*)

Calcification in Twins.—Pulmonary calcification in ten-year-old twin sisters is reported. Their skin tests were negative with tuberculin and coccidioidin, but positive with histoplasmin. They had lived in Nebraska, Colorado, Illinois, Kansas, California and Michigan, several of which are in the Mississippi basin area where histoplasmosis is believed to be most prevalent. The child with the greater amount of calcification had previously had asthma, had a low grade fever, but had no current lung symptoms. Studies of sputum and nodes produced no fungus.—*Pulmonary calcification in twins, G. Houston & W. A. Steiger, J. Pediat., June, 1948, 32: 706.*—(*W. H. Oatway, Jr.*)

Streptomycin for Tuberculous Meningitis.—The results of treatment of 72 cases of tuberculous meningitis are recorded; 13 of these began as miliary tuberculosis. In 9 cases no streptomycin was given and death ensued. The drug was given in 63 cases, of whom 34

died. It was administered both intramuscularly and intrathecally. The dosage was 1.0 Gm. daily or less. The more severe the signs, the smaller should be the intrathecal dose. The average duration of treatment was three to four months. Continuous treatment was given initially for four to six weeks, resulting in amelioration of signs and symptoms, but in persistence of the laboratory evidence of infection. After a lapse of five to ten days, treatment was resumed for ten to fifteen days. This schedule of intermittent treatment was continued until there were no laboratory signs of infection. As long as the cell count and chemical findings remained abnormal, the dangers of exacerbation were great. By lengthening the duration of the disease and prolonging life, the treatment increased the incidence of hydrocephalus. The irritating effects of large doses of streptomycin contributed to the development of this complication. Thus, in the series treated with streptomycin more of the fatalities were due to hydrocephalus than in previously untreated cases. There is less risk of hydrocephalus in cases in which treatment is begun early. Of 50 treated cases of tuberculous meningitis, 21 were cured, 3 are still under treatment and 26 died. The survivors were usually the ones in whom treatment was started in less than ten to fifteen days after the onset of the disease. In the fatal cases, the length of survival was longer than in untreated cases. In 13 cases of miliary tuberculosis complicated by meningitis, there were cures in 2, relapses in 2, improvement in one and death in 8 cases. These cases require larger doses and longer treatment. It is believed that paratracheal adenitis in primary tuberculosis is often a forerunner of meningitis; in these cases the use of streptomycin prophylactically is recommended.—*Streptomycin treatment of tuberculous meningitis in children, K. Choremis, N. Zerros, V. Constantinides & S. Pantazis, Lancet, October 16, 1948, 2: 595.*—(*A. G. Cohen*)

Streptomycin for Meningitis.—Nine patients ranging from 18 months to 35 years of age were treated with streptomycin for tuberculous meningitis. Three of the cases showed a notably good response, although one is blind and the other 2 have not been followed long; the other 6 died. Four of the patients had miliary tuberculosis of the lungs, 2 had chronic pulmonary tuberculosis (one with genito-urinary lesions), one had a tuberculous pleural effusion, and 2 had calcifications in the lungs or spleen. Therapy consisted of intramuscular injections every six hours; the daily dosage ranged from 0.25 Gm in infants to 4 to 8 Gm in adults. Small intrathecal doses were sometimes used. Toxic reactions from the drug were common and often severe. The failure of many brain lesions to respond to streptomycin is probably due to their inaccessibility. These results are similar to those in other reports.—*The treatment of tuberculous meningitis, L. Alperin & J. A. Toomey, J. Pediat., July, 1948 33: 74.*—(*W. H. Oatway, Jr.*).

Tuberculous Mastoiditis.—An infant aged eight weeks was found to have tuberculous mastoiditis. Following a cortical mastoidectomy, streptomycin was administered for fourteen weeks. The treatment resulted in a complete cure. This is noteworthy in view of the previously grave prognosis in such cases; moreover, a less radical surgical procedure was sufficient.—*Tuberculous mastoiditis, O. D. Fisher & E. A. Malkin, Lancet, October 30, 1948, 2: 689.*—(*A. G. Cohen*)

Streptomycin for Pulmonary Tuberculosis.—Good results were obtained in cases of bronchial and laryngeal tuberculosis and tuberculous sinuses of the chest wall. Results were good in cases of tuberculous pneumonia but disappointing in one case of exudative tuberculosis.—*Streptomycin in pulmonary tuberculosis, R. Y. Keers, Lancet, September 18, 1948, 2: 449.*—(*A. G. Cohen*)

Subtilin for Tuberculous Lesions.—Subtilin was administered intraorally by a nebulizer to 8 patients ranging from 18 to 60 years of age. Four of the patients had tuberculous

laryngitis, one had endobronchial disease and the remaining three were considered to have a conspicuous bronchial factor in their pulmonary disease although lesions were not observed in the main stem bronchi at bronchoscopic examination. Subtilin therapy in the latter was directed toward presumed endobronchial lesions in the smaller bronchi. The progress of the disease was followed during therapy by means of frequent physical, fluoroscopic, and roentgenologic examinations. To determine the action and possible toxicity of subtilin, the following laboratory procedures were performed: determination of vital capacity, measurement of daily sputum output, urinalysis, complete blood cell count, sedimentation rate, nonprotein nitrogen and sugar in the blood, protein in the serum, albumin-globulin ratio, icterus index, cephalin flocculation, bromsulfalein liver function test, and phenolsulfonphthalein renal function test. The sensitivity of the tubercle bacilli to the antimicrobial action of subtilin was determined in 6 cases prior to treatment. Of the 4 patients with tuberculous laryngitis, improvement was marked in one, slight in one and questionable in one. In the fourth the disease progressed and the patient died. In the one subject with endobronchial disease, which could be carefully evaluated because of extensive lesions visible at bronchoscopy, it appeared that subtilin favorably influenced the endobronchial disease. After eight and one-half months of therapy bronchoscopy revealed no occlusion or other evidence of endobronchial disease. After ten months of subtilin therapy another bronchoscopic examination revealed inflammation and granulations involving the pharynx, larynx, trachea and bronchi. Tubercle bacilli cultured from the sputum at this time were twice as resistant to subtilin as organisms recovered before treatment. Although the development of marked bacterial resistance was suspected, this was not proved by cultural studies, and the cause of the reactivation of the endobronchial disease is unknown. The condition in the 3 patients in whom endobronchial tuberculosis was presumed to be present in the small bronchi showed no significant change during therapy. Suggestive evidence of the development of bacterial resistance to subtilin *in vivo* was obtained in 3 cases. No evidence of toxicity in the kidneys, liver or bone marrow was encountered. The lack of serious toxicity encountered may be inherent in the mode of administration which results in negligible systemic absorption of subtilin. Administration of subtilin by inhalation was associated with irritation of the respiratory tree in almost all patients during the first two or three weeks, after which the symptoms subsided or disappeared. Asthma developed in one subject and the drug was discontinued. Further investigation of the therapeutic possibilities of subtilin must await basic research directed towards development of derivatives more soluble in physiological fluids. Subtilin in its present form has no place in the treatment of tuberculosis other than for purely investigative purposes.—*Topical application of subtilin to tuberculous lesions in man, S. M. Farber, H. R. Eagle, H. H. Anderson, & R. D. Gorman, J. Lab. and Clin. Med., July, 1948, 33: 799.*—(*F. G. Petrik*)

Prevention of Pulmonary Infection.—Chronic pyogenic bronchopulmonary disease results from primary acute infections of several types. It is essential that the acute conditions be treated vigorously until cured and also that chronic purulent bronchitis be controlled. A study of 50 infants and children who had been treated in the hospital with sulfonamides and/or parenteral penicillin showed many to have had either ill health and progressive pulmonary disease or good health and recurrent respiratory infections in the ensuing year. Pulmonary emphysema and fibrosis often accompanied the chronic lesions. Children with acute or residual infections should be checked at intervals, as are those who have had tuberculosis or rheumatic fever. In those with acute, subacute, or chronic infections, penicillin aerosol therapy may be of value. It may require weeks or months of daily treatments to heal or stabilize the condition and recurrent treatment may be needed. A plastic

nebulizer has been used; a bicycle pump is more convenient and less expensive for prolonged use than oxygen pressure or a hand-bulb. Anti-histaminic drugs alleviate oral sensitivity. Resistant organisms have not developed and sputa have shown only susceptible bacteria. Failure to respond apparently has been due to the serious character of the disease or to an improper therapeutic technique. Best results are obtained in the more acute conditions but hope can be offered the chronic cases. Therapy also removes a public health hazard.—*Prospects for prevention of chronic bronchitis and bronchiectosis, W. Finke, J. Pediat., July, 1948, 33:29.—(W. H. Oatway, Jr.)*

Postoperative Pulmonary Complications.—Patients about to undergo abdominal operations were given thorough chest examinations on the basis of which they were graded as good, moderate or bad risks. Alternate cases in each of the groups received 3 inhalation treatments consisting of 100,000 units of nebulised penicillin on the day before operation, and 2 additional treatments on the following day. Bacteriological studies showed considerable diminution in the number of penicillin-sensitive bacteria in the nasopharynx. However, the incidence of postoperative pulmonary complications was not reduced.—*Nebulised penicillin and postoperative pulmonary complications, E. J. Holborrow & E. A. Spriggs, Lancet, October 30, 1948, 2:488.—(A. G. Cohen)*

Effect of Medium on M. Tuberculosis.—The effect of Tween 80, bovine albumin, glycerol and glucose on the growth of *M. tuberculosis* H37Rv was evaluated by using nitrogen determinations to determine the amount of growth of tubercle bacilli. The media employed were a modified Proskauer and Beck synthetic medium and a medium recommended by Dubos. The results show that unpurified Tween 80 (0.05 per cent) exerted an inhibitory effect in both media and that purified Tween 80 exerted a similar bacteriostatic effect but to a lesser degree. Both glycerol and glucose markedly stimulate growth. Purified Tween 80 also stimulates growth when added to either of the media in the absence of glucose or glycerol, but not to nearly the same degree as either glucose or glycerol alone. Purified Tween 80 when added to either of the media in the presence of glycerol or glucose inhibited growth as compared with either glucose or glycerol alone. This is in contrast to the findings of Dubos and his collaborators that glycerol inhibited the growth of small inocula in the presence of Tween 80. Bovine serum albumin (fraction V; 0.2 per cent) did not stimulate growth of the tubercle bacillus; it protected the organism against the inhibitory effect of purified and unpurified Tween 80 (0.05 per cent) but only during the first five days of growth. The modified Proskauer and Beck medium containing 2.0 per cent glycerol supported growth of tubercle bacilli at a maximum rate for a longer period than the Dubos medium containing 0.2 per cent glucose.—*The effect of "Tween 80", bovine albumin, glycerol, and glucose on the growth of Mycobacterium tuberculosis var. Hominis (H37Rv), T. H. Sattler & G. P. Youmans, J. Bact., August, 1948, 56: 235.—(F. G. Petrik)*

Effect of Tween 80 on Tuberculostasis.—It has been reported that the presence of Tween 80 in synthetic media increased the bacteriostatic action of penicillin, streptomycin, and subtilin on the subsurface growth of tubercle bacilli. Other investigators found, on the contrary, that Tween 80 reduced the inhibitory action of certain compounds on the growth of this organism. As a result of these contradictory reports, this work was undertaken to determine what effect Tween 80 would have on the *in vitro* bacteriostatic activity of 20 compounds for *M. tuberculosis var. hominis* H37Rv. The results show that Tween 80 added to Youman's basal medium without albumin increased the tuberculostatic activity of 15 of 20 compounds, decreased the activity of 3 compounds and had no effect on the action of 2 compounds. The addition of bovine albumin alone to the basal medium reduced

the bacteriostatic action of 13 of the 20 compounds but did not increase the degree of bacteriostasis of any. In the medium with Tween and albumin, 5 compounds were more bacteriostatic than in the basal medium, 4 gave approximately the same results in the two media, and the remaining 11 were less bacteriostatic than in the basal medium. Unpurified and purified Tween 80 gave approximately the same results with 7 of 14 compounds and in the remaining 7 compounds purified Tween 80 produced less inhibition of bacteriostasis than the unpurified detergent. Under the conditions of the experiments the free oleic acid that may be present in Tween 80 was not responsible for the effect of this compound on bacteriostatic activities. In the presence of albumin the bacteriostatic activity of 12 of the 13 compounds was essentially the same with purified and unpurified Tween 80. The explanation for the divergent results obtained with Tween 80 on the 20 compounds employed in this study as compared with the results of other investigators is not apparent. There appears to be no relationship between the action of Tween 80 on these compounds and their chemical structures nor did a fourfold reduction in the concentration of Tween 80 produce significant changes in the results with 5 compounds. The irregular results produced by the presence of Tween 80 on the tuberculostatic activity of compounds indicate the desirability of employing as simple a synthetic medium as possible. Plasma, serum, or albumin can be added to the synthetic medium as a separate test to determine the inactivation of the compound by protein.—*The effect of "Tween 80" in vitro on the bacteriostatic activity of twenty compounds for Mycobacterium tuberculosis, A. S. Youmans & G. P. Youmans, J. Bact., August, 1948, 56: 245.—(F. G. Petrik)*

Resistance of Mycobacteria to Streptomycin.—This is a report of studies made of the orderly manner in which spontaneous variants occur in cultures of *M. tuberculosis* and *M. ranae*. The strains of tubercle bacilli used were H37Rv and 2 strains, WS and WR, isolated from the sputa of patients prior to the institution of treatment with streptomycin, one (WS) becoming resistant and the other (WR) remaining sensitive to streptomycin after treatment. Liquid and solid Tween-albumin media were employed. The results show that when large samples of bacteria were seeded on media containing streptomycin, resistant forms were isolated, the number being directly affected by the concentration of the drug in the media. A marked decrease in the number of colonies occurred when the drug concentrations were increased from 1 γ to 10 γ per ml. and then to 100 γ per ml. When certain high concentrations of the drug were used, such as 100 and 1,000 γ per ml., no significant difference in the number of colonies was observed. More resistant variants were found in the strain WS, which later developed resistance to streptomycin after thirty days' treatment, than in WR which remained sensitive to streptomycin. When the sample of bacteria used was sufficiently large and when the time allowed for growth was prolonged, the chances of finding resistant forms were increased. When only one sample is used, the conclusions drawn must be very limited, for great variations were noted among samples from the same parent population. When sensitivity studies are made in the usual manner, a time limit is used for allowing growth of resistant forms. When the time limit is extended, a few resistant forms may be allowed to multiply adequately and the results may be given a quite different interpretation. In addition, the collection of sputum specimens from which the original stock cultures are grown must be considered inadequate in certain instances. All the varitions in resistance of the bacterial population in the diseased areas of the patient are probably never represented in any single or even in several sputum samples. It is conceivable that all the bacteria within a patient are not exposed to the same concentration of streptomycin. Because of the low concentration of streptomycin in certain of the diseased areas, bacteria which may be readily susceptible to streptomycin are allowed to exist during the treatment period and it is con-

ceivable also that an extension of disease in the patient after the treatment period is concluded could be caused by these streptomycin-susceptible organisms. It is essential that complete studies be made of tubercle bacilli obtained from many different areas in patients who have undergone treatment with streptomycin and subsequently have died. It is known that resistance is specific and permanent in nature. Is it not possible that resistant variants may occasionally give rise to mutants resembling the parent strain in their susceptibility to streptomycin? From large populations of a variant of *M. ranae*, which requires streptomycin for growth, a few colonies have been isolated that resemble the parent strain in their susceptibility to streptomycin and sulfonamides, in their biochemical reactions, and in their morphological and cultural characteristics.—*A quantitative analysis of the resistance of Mycobacteria to streptomycin, D. Yegian & R. J. Vanderlinde, J. Bact., August, 1948, 56: 177.*—(*F. G. Petrik*)

Streptomycin-Resistant Variants.—Streptomycin-resistant variants were obtained from 25 of 57 recently isolated strains of tubercle bacilli. This fact suggests that these variants may play a role in the development of the streptomycin resistance which occurs in cultures of tubercle bacilli isolated from patients who have had prolonged treatment with streptomycin.—*Streptomycin resistant variants obtained from recently isolated cultures of tubercle bacilli, G. P. Youmans & E. H. Williston, Proc. Soc. Exper. Biol. & Med., July, 1948, 68: 458.*—(*F. B. Seibert*)

Streptomycin and Tuberculosis of Mice.—Streptomycin to the amount of 3,000 γ per day had a favorable effect upon the course of experimental tuberculosis in mice when treatment was initiated seven, fourteen and twenty-one days following infection. All treated mice eventually died and the extent of the pulmonary disease did not differ significantly from the control animals. They all exhibited proliferative lesions, whereas the lesions of the controls were predominantly necrotic exudative. The lesions in the treated animals were extensive and occupied almost all alveoli, but the numbers of tubercle bacilli were considerably reduced as compared with the controls.—*The effect of streptomycin on well established experimental tuberculosis of mice, G. P. Youmans, E. H. Williston, A. S. Youmans, & R. R. Osborne, Proc. Soc. Exper. Biol. & Med., August, 1948, 68: 661.*—(*F. B. Seibert*)

Dietary Lipids and Experimental Tuberculosis.—Administration of a nonlipid, casein-supplemented ration to resistant Swiss albino mice retarded the progress of experimental tuberculosis. Retardation also occurred on a ration of which 20 per cent was the total fatty acids of cocoanut oil. Olive oil, linseed oil, and oleic acid in a concentration of 20 per cent enhanced the progress of experimental tuberculosis.—*Influence of dietary lipids on experimental tuberculosis, L. W. Hedgecock, Proc. Soc. Exper. Biol. & Med., May, 1948, 68: 106.*—(*F. B. Seibert*)

Tuberculosis in the Rabbit Ear Chamber.—Microscopic studies of the tissue changes in developing tuberculous infection within a Clark chamber were made. The early reaction to the presence of living tubercle bacilli was minimal. The late reaction was an explosive, necrotizing response. In the one rabbit systematically skin tested with Old Tuberculin this late reaction began coincidentally with the development of skin hypersensitivity. Progressive vascular damage ending in venous thrombosis played an important role in tissue destruction. The bovine tubercle bacillus (Strain Ravenel RV) was used.—*Development of tuberculous infection: In vivo observations in the rabbit ear chamber, R. H. Ebert, J. J. Ahern & R. B. Bloch, Proc. Soc. Exper. Biol. & Med., August, 1948, 68: 625.*—(*F. B. Seibert*)

Mouse Test for Tuberculosis.—Gastric mucin significantly enhances the virulence of tubercle bacilli for pigmented strains of mice. Grossly visible bacilli are usually present ten days after intraperitoneal inoculation with a dose of about 500,000 bacilli of the H37Rv

strain. Several isolations of tubercle bacilli from patients have been made ten to fifteen days after the inoculation of pigmented mice with mixtures of sputum and mucin. Recently the number of isolations from patients was increased following inoculation of pigmented mice which had been fed the cornmeal-gelatin butter diet of Dubos and Pierce.—*A rapid mouse test for laboratory diagnosis of tuberculosis, A. Milzer & E. R. Levine, Proc. Soc. Exper. Biol. & Med., October, 1948, 69: 16.*—(*F. B. Seibert*)

Tubercle Bacilli in Cerebrospinal Fluid.—In 2 cases of primary tuberculosis in infants, tubercle bacilli were isolated from the cerebrospinal fluid. In neither case was there clinical or chemical evidence of meningitis. One patient was given streptomycin; both patients survived. The source of the bacilli was felt to be a small meningeal lesion which subsequently healed.—*Primary tuberculosis with meningism and bacilli in the spinal fluid, K. Choremis & G. Vrachnos, Lancet, September 11, 1948, 2: 408.*—(*A. G. Cohen*)

Passive Transfer of Hypersensitivity to Tuberculin.—The method of local passive sensitization so extensively employed in human allergy (Prausnitz-Küster reaction) and in experimental anaphylaxis (Opie and others) can also be applied to the cellular transfer of tuberculin hypersensitivity discovered by Chase. Cells from a peritoneal exudate of a normal guinea pig were injected intracutaneously into the skin of the left flank and cells from a peritoneal exudate of a tuberculous guinea pig were injected into the right flank of a normal guinea pig. After forty-eight hours 0.25 cc. Old Tuberculin with 0.25 cc. saline was given subcutaneously, and fifteen minutes later 1.25 cc. Old Tuberculin with 1.25 cc. saline were given intraperitoneally. Immediate shock occurred followed by complete recovery in a few minutes. Twenty-four hours later a definite reaction occurred in the skin site prepared with "tuberculous" cells but not in the skin of the left flank.—*Passive transfer of local cutaneous hypersensitivity to tuberculin, M. N. Metaxas & M. Metaxas-Bühler, Proc. Soc. Exper. Biol. & Med., October, 1948, 69: 163.*—(*F. B. Seibert*)

Experimental "Wet Lung".—The accumulation of blood, transudates, exudates, or mucus in the lungs following accidental or operative trauma is termed "wet lung" or, loosely, "pulmonary edema." It is a hazard to recovery and the mechanism of production has been poorly understood. A series of experiments has been done to study the problem from the traumatic and neurogenic aspects. Anesthetized dogs were subjected to measured blunt and tearing trauma to the chest wall; "wet lung" was produced. The condition was present not only near the site of injury but often was widespread. The condition could be aggravated by infusions of isotonic saline solution which were harmless in normal animals. Reduction of oxygen intake plus saline infusions also produced "wet lung". Measurement of various blood elements and dynamics did not reveal reasons for the widespread nature of the edema following localized injury. Dorsal sympathectomy afforded marked protection against trauma and infusions, and bilateral cervical vagotomy was also somewhat protective. This suggests that generalized "wetness" is in part a reflex phenomenon.—*"Wet lung"—An experimental study, R. A. Daniel Jr. & W. R. Cate, Jr., Ann. Surg., May, 1948, 127: 830.*—(*W. H. Oatway Jr.*)

Silicosis.—The essential agent in the causation of true or "classical" silicosis is the freshly cloven silica particle. The pathological process basically consists of the hydration of the silica particle at the expense of the cell protoplasm. Because of its atomic lattice structure, quartzite when powdered yields a more virulent dust than other silica formations. Fully hydrated silica (silica hydrosol, sometimes mistakenly called "silicic acid") is non-toxic and enters freely into the metabolism of plants and animals. Partly hydrated suspensions of silica in water ("colloidal silica") retain some of the chemical activity of the dry powder but to a much less degree.—*What is*

silicosis, P. Heffernan, Tubercle, August, 1948, 29: 169.—(A. G. Cohen)

Tuberculosis in Adolescents.—While medical supervision of the school population is good, this is not true of youths after they leave school. The advent of puberty makes these young people peculiarly susceptible to tuberculosis. Three external circumstances combine to add to the hazards of the immediate post-school period: (1) the greater mental and bodily strain of professional life; (2) leaving the parental home for living conditions which may be less favorable; and (3) greater opportunity for infection through wider contact. All forms of childhood and adult tuberculosis are seen in these young people and there is nothing characteristic about the character or the course of the infection. Contrary to common belief, primary tuberculosis heals about as readily in these people as it does in children. Reinfection tuberculosis shows very rapid progression at times, occasionally in the presence of a healing primary focus. The rapid progression from initial focus to advanced disease makes it clear that periodic roentgenographic examination is not the whole answer to early detection, and supervision by the family physician is essential. Two laws are before the legislature that deal with the problem of tuberculosis in adolescents. One authorizes periodic roentgenograms of groups of people at special risk, and the other law deals with compulsory BCG vaccination of all young people who are tuberculin-negative when leaving school. Among 40 cases analyzed, it appeared that 17 were the result of recent primary infection, so that BCG vaccination may presumably be of benefit in somewhat less than half of recognized cases in young adults.—*Die Tuberkulose der Schulentlassenen, M. Wissler & M. Meier, Schweiz. med. Wchnschr., December 27, 1947, 77: 1349.—(M. Marcus)*

Tuberculin Testing of Army Recruits.—Army recruits around the age of twenty were tuberculin tested. Mantoux tests were done at a dilution of 1:1,000 of a solution containing the products of both human and bovine bacilli. The negative reactors were given another test with the same material in 1:100 dilution. It was found that in a group of 1,902 young men, only 20 per cent were negative reactors. The actual figure of negative reactors of this age group is even lower because all patients under treatment were excluded, and because preceding roentgenograms eliminated all those with asymptomatic pulmonary lesions judged to be tuberculous. The figures are in sharp contrast with those obtained elsewhere, notably in France. There a survey conducted in 1946 among army inductees showed that 52 per cent of 100,000 men were tuberculin negative. One may only speculate as to the causes for the discrepancy of results. The author feels that the fact that his antigen also contained products of bovine bacilli may be of significance.—*Premières données des intradermo-réactions tuberculiniques pratiquées chez les recrues suisses, E. Delachaux, Schweiz. med. Wchnschr., December 18, 1947, 77: 1304.—(M. Marcus)*

CHEMOTHERAPEUTIC MEASURES IN TUBERCULOSIS: GENERAL ASSESSMENT AND CURRENT PROBLEMS[1]

P. D'ARCY HART[2]

HISTORICAL BACKGROUND

The ever widening field in which chemotherapy has achieved clinical success leaves the hitherto resistant infective diseases in increasing prominence. Among these, tuberculosis stands out as the main killing disease of bacterial nature affecting the productive groups of the population. The door to success at last seems to be opening, however, for in the past ten years clinical evidence of the value of a few substances has appeared. A number of others show promise. Let us, by way of introduction, take a brief look at the lines along which some of the researches have been traveling.

Present day developments with synthetic chemical agents date from the observation in 1938 by Rich and Follis (1) in Baltimore, that experimental tuberculosis in the guinea pig could be retarded by sulfanilamide. Trial of this sulfonamide in tuberculosis was a logical consequence of the historic discovery in Europe four years previously that the drug exerted an appreciable effect in acute bacterial infections. Workers at the Pasteur Institute in Paris (2, 3) soon demonstrated the more impressive anti-tuberculous activity of the related sulfone compounds, and this series has been developed and exploited in many countries since 1939.

Effective study of therapy with antibiotic substances, *i.e.*, with antimicrobial substances produced by species of living microorganisms, dates only from the introduction of penicillin as a chemotherapeutic agent in 1940. This great advance, by encouraging intensive study of other soil microorganisms, led Waksman and his colleagues (4) in New Jersey to the isolation of streptomycin in 1944. This drug has been given more experimental and clinical trial in tuberculosis than has any other product of microbial origin. A recent general bibliography of streptomycin (5) lists no less than 263 papers having a bearing on tuberculosis.

The development of the use of calciferol (vitamin D_2) in massive doses in lupus vulgaris has historical links with the treatment with enormous doses of cod liver oil given a century ago for many kinds of tuberculosis and for lupus (6). Moreover, the use of calciferol is linked also with heliotherapy in lupus insofar as this stimulus marshals the body's resources of vitamin D. Treatment with calciferol was introduced independently, and perhaps empirically, during World War II by Charpy (7) in France and by Dowling and Thomas (8) in England. Calciferol is not antibacterial in the sense of most chemotherapeutic agents but probably acts on the tissues of the host.

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Quite different was the mode of origin of the use of the synthetic substance para-aminosalicylic acid (PAS) which was introduced in Sweden in 1946 by Lehmann (9). The development of the drug resulted from knowledge of the metabolism of the tubercle bacillus that had accumulated during the previous decade, notably as a result of Bernheim's (10, 11) work in America.

These four chemotherapeutic agents or groups of agents illustrate both the diversity of approach of research to this problem and, in a wider sense, the internationality of medical science. With their mention we have all but covered the few substances that have already shown evidence of usefulness in tuberculosis, or are "in the running." I shall return to their assessment later.

RECENTLY REPORTED SUBSTANCES ACTIVE AGAINST MYCOBACTERIUM TUBERCULOSIS

In 1946 I had occasion (12) to tabulate the various substances reported to be active against the tubercle bacillus in the test tube or against tuberculous infection in the animal body. During the past two years the list has grown formidably. There is no need to present a survey of the subject in full, in view of several recent authoritative reviews (13 to 17), but we must consider what has been the positive yield from all these researches. Most of the substances studied have not been tested for antituberculous activity except in the test tube. Some have proved to be too toxic; others are inactivated in the body; and still others, particularly the antibiotics, may not yet have reached a stage suitable for animal experimentation because of problems related to purification.

In table 1 may be seen a summary of most of the substances or groups of substances reported to have some degree of antituberculous activity on the basis of recent tests in experimental tuberculous infection in animals, or in clinical tuberculosis in man.

Examples will be noted of purely synthetic substances, *e.g.*, promizole at one extreme, and products of biological origin, *e.g.*, streptomycin, at the other. The successes in animal experimentation number not much more than a score, those claimed or proved in man, half a dozen. An attempt will be made to assess these latter individually and very briefly.

ASSESSMENT OF CHEMOTHERAPEUTIC SUCCESSES CLAIMED IN MAN

Several hundred *sulfone-compounds* have been tested in the laboratory and the more promising of these have been studied in man. The latter include two of rather low toxicity, promizole (12 and 21), and the recently introduced sulfetrone (22, 77). It is a fair statement to say that the clinical results, while indicating some regressive effect, *e.g.*, in acute miliary tuberculosis (27) and in certain types of pulmonary tuberculosis (28–30), have not so far fulfilled the promise of the animal experiments *when these agents have been used alone.* Hinshaw and Feldman's (78) statement in 1945 that, as sole medicaments, "no definite place has been found for these drugs in treatment of the usual types of tuberculosis" seems still to be true. In combination with streptomycin therapy, however, the outlook is considerably more promising. Encouraging results have been reported in meningitis cases treated with promizole and

TABLE 1

Substances reported as antituberculous in vivo (1938 to 1948)

EXPERIMENTAL ANIMALS		MAN
(a) *Sulfonamides and sulfone compounds*		
Sulfanilamide (1, 18 and see 19)		
Other sulfonamides (20)		
Promin (see 12, 19, 21)		
Diazone (see 12 and 21)		
Promizole (see 12 and 21)		Promizole (26, 27; and see 12 and 21)
Sulfetrone (22)		Sulfetrone (28 to 31)
Other sulfones (23 to 25; and see 12)		Other sulfones (32, 33)
(b) *Fatty acids and derivatives*		
Ethyl palmitate and stearate (34)		
Certain chaulmoogra derivatives (35)		
(c) *Aromatic compounds*		
Certain naphthoquinone derivatives (36, 37)		
1-diethylamino 2-(2,4,6-triiodophenoxy)-ethane HCl (38)		
Para-aminosalicylic acid (39 to 43)		*Para*-aminosalicylic acid (44 to 47)
Thymol (48, 49)		
1,1,1-trichloro-2,2-bis (*P*-aminophenyl)-ethane (50, 51)		
2-butoxy-6-aminobenzothiazole HCl (52)		
Certain diaminomethylpyrimidines and related compounds (53, 54)		
(d) *Miscellaneous organic compounds*		
Nicotinamide (104, 105)		
Amido-compounds of α-furancarbonic acid (55)		
P-acetamidobenzaldehyde thiosemicarbazone (56)		*P*-acetamidobenzaldehyde thiosemicarbazone (106)
Ergosterol and related compounds (57, 58)*		Calciferol (7, 8, 59 to 63)
(e) *Antibiotics, etc.*		
Streptomycin (see 17, 21, 64)	Actinomycetes	Streptomycin (66 to 70, 100)
Mannosidostreptomycin (65)	Actinomycetes	
Chloromycetin (102)	Actinomycetes	
Subtilin (71, but see 72 and 107)	Bacteria	
Licheniformin (73)	Bacteria	
Nisin (74)	Bacteria	
Cepheranthine (see 75)*	Higher plants	Cepheranthine (see 75)
Usnic acid (76)	Higher plants	

* W. H. Feldman and A. G. Karlson, in unpublished studies, have failed to confirm the observations of others that either ergosterol (nonactivated) or cepheranthine is an antituberculous agent in experimentally infected guinea pigs (Personal communication, W. H. Feldman).

streptomycin simultaneously (26) and further trials of combined therapy are in progress in many countries.

Clinical trials of *para-aminosalicylic acid* (*PAS*) have been in progress in Sweden for two years (44, 47) and have been recently started in England (45, 46), in the United States and elsewhere. Yet, from reading the few papers so far published it is difficult to come to a firm conclusion on the efficacy of this drug in human pulmonary tuberculosis, or even in tuberculous empyema where apparently dramatic results may occur. Reports on the use of the drug in the treatment of meningitis are meagre. PAS is difficult and expensive to manufacture, and the early preparations were less satisfactory and more toxic than some in use at present. With the increased availability of satisfactory preparations of the drug, the wider trials now taking place should give an answer, provided they are properly designed. The administration of PAS, both alone and in combination with streptomycin, has been followed by significant changes in the course of experimental tuberculosis in the hands of some workers (23, 39, 42, 43), though not of all (41). There is no doubt that this interesting substance requires thorough examination in the field.

The dramatic and consistent improvements taking place under *calciferol* therapy (62) leave no doubt as to its efficacy in lupus vulgaris and certain other forms of skin tuberculosis. In tuberculous adenitis and possibly other non-pulmonary tuberculous conditions, suggestive results have also been obtained though the benefit is not clearly proved. In pulmonary tuberculosis the evidence for the efficacy of calciferol is very doubtful.

The Japanese (see 75) have claimed that the administration of minute doses of the alkaloid *cepheranthine* has resulted in improvement in many types of clinical tuberculosis. This drug is mentioned here only because the claims are so strong although well documented evidence is still awaited.

The discovery of *streptomycin* was of historic importance, not only because it was tuberculostatic in the test-tube, for many other substances have this property though not often to the same degree, but because of its low toxicity which permitted its use in effective doses in animals and man. The successful results observed with the streptomycin treatment of experimental tuberculous infections in guinea pigs (79) heralded the first real advance in the long struggle against this disease. Moreover, these results revealed the error of the ideas previously held by many that, in order for the chemotherapy of tuberculosis to be successful, it would be necessary to overcome the obstacle of the supposed fatty coat of the tubercle bacillus (streptomycin is water-soluble yet apparently passes readily into the cell).

The present status of streptomycin therapy in human tuberculosis has been succinctly described in various American reports, including that of the American Trudeau Society (66 to 69, 80). It is sufficient to state here that British Medical Research Council trials confirm the immediate life saving power of this antibiotic in a proportion of cases of tuberculous meningitis (70, 81) and acute miliary tuberculosis. Like the American workers, we have also found that streptomycin retards further progression and causes rather swift regression of

that element of pulmonary tuberculosis that is variously called infiltrative, exudative or bronchopneumonic. The striking improvements in some cases of ulcerative tuberculous laryngitis and tracheobronchitis following streptomycin therapy are sufficiently well known to require no further comment.[3]

SOME DEFECTS OF PRESENT ANTITUBERCULOUS AGENTS

Consideration will now be given to certain failures to meet the requirements for a satisfactory chemotherapeutic agent revealed by clinical trial of the various antituberculous substances.

A most important defect is toxicity. While the toxicity of streptomycin is remarkably low when considered in relation to effective dose, it nevertheless constitutes a problem. The permanent damage to the vestibular apparatus, even though it may be compensated for to some extent, provides a sound reason for the view that streptomycin is at present usually contraindicated in minimal pulmonary tuberculosis and in those other forms of the disease which may be satisfactorily treated by other measures. PAS can be given by mouth and seems to have little toxicity in man in the dosage used at present, but its efficacy is still *sub judice*. The sulfone drugs produce toxic blood changes to a varying degree, though these are comparatively slight with some of the newer products, particularly when administered by the parenteral route. The goitrogenic activity of the compounds, though again slight in some, is a distinct disadvantage. Massive doses of calciferol not uncommonly produce toxic effects, but these are seldom of sufficient severity to contraindicate the use of the drug.

Another and notorious bugbear of certain antituberculous agents, particularly streptomycin, is the phenomenon of the appearance of drug-resistant strains of bacilli. In the case of streptomycin, drug-resistant strains are frequently demonstrable after a month or more of treatment. It is of interest that this phenomenon seems to occur much less frequently in the course of meningitis than in pulmonary tuberculosis. If the great majority of the infecting bacilli cease to be susceptible to the drug, the curve of the patient's progress may flatten out after an initial improvement. Likewise, a patient may do well at first under streptomycin therapy but, if the disease flares up, say a year or more later, it may then be insusceptible to the drug. Consequently another reason against indiscriminate use of streptomycin in minimal disease amenable to ordinary treatment is the fear of spoiling the patient's chances for successful chemotherapy in the event of a subsequent relapse. From the community standpoint there is also a danger that new cases of tuberculosis will arise from contact with persons previously treated with streptomycin and who are discharging drug-resistant bacilli. The bacilli in such secondary cases might be

[3] A further group of compounds should be included under the heading or recent chemotherapeutic successes claimed in man. With the sulfonamides as starting point, Domagk and his collaborators in Germany have branched off on a new line, resulting in active thiosemicarbazones (56). Two of these, *p*-acetamidobenzaldehyde thiosemicarbazone and anisaldehyde thiosemicarbazone, have been given clinical trial, benefit in lupus vulgaris being claimed (106).

streptomycin-resistant from the very beginning of the infection and hence the disease would not be affected by streptomycin therapy.

The toxicity problem is being attacked in a number of ways. The most obvious approach is to synthesize new substances and to continue the search for fresh antibiotics. Attempts are being made to alter the chemical constitution of certain of the antituberculous antibiotics, including those that have not progressed beyond animal experimentation, in the hope of reducing toxicity more than antimicrobial activity.[4] It is possible that the same end may eventually be achieved by the synthesis of analogues of the simpler of the antibiotics. Reduction of the toxic effects of streptomycin is being attempted in the field by various modifications of the dosage schedule, as in current trials sponsored by the National Institute of Health and by the use of streptomycin in conjunction with other drugs, *e.g.*, the sulfones or PAS, in order to reduce the required dosage of the antibiotic. The results of animal experimentation (23, 43, 82, 83 and see 73 and 84) suggest that both of these lines of investigation have promise.

Various approaches are also being explored towards prevention of the baffling phenomenon of the emergence of drug-resistant strains of bacteria under streptomycin treatment. The attempts include variations in the schedules of administration to see if the emergence to predominance of drug-resistant bacilli can be delayed or prevented. The administration of streptomycin in conjunction with other chemotherapeutic agents may influence the phenomenon of drug-resistance, although a few results of simultaneous clinical use of sulfetrone and streptomycin (31) have not been encouraging. It would indeed be useful to have another drug to bring into action after the emergence of bacilli resistant to streptomycin. On the other hand, the fact that, in our experience, some cases of pulmonary tuberculosis continue to improve, even when resistance to streptomycin has apparently developed, may indicate that an important proportion of the bacilli are still sensitive (85). It is equally possible, however, that such improvement represents an effect of the patient's own resistance after the action of the streptomycin has ceased (86).

Defects other than those just mentioned are associated with one or more of the antituberculous agents under investigation at present. Caseous tissue seems to resist penetration by streptomycin, and pus is an unfavourable medium for its action, possibly on account of the depressive effect of an acid pH on an antibacterial drug of basic nature (87). This phenomenon may explain why the effects of streptomycin tend to be limited to the "exudative", bronchopneumonic elements of tuberculosis rather than the more chronic caseous elements, and why the drug is notably ineffective in tuberculous empyema. There is the possibility that PAS an acidic drug, does not possess these disadvantages.

In summary, it must be admitted that no single agent, not even streptomycin, has yet satisfied all the requirements for full chemotherapeutic success. The most important of these requirements are (88): (1) absence of serious or irreversible physiological derangements; (2) reversal of established progressive disease to nonprogression, resolution, fibrosis or calcification; (3) results to be

[4] This appears to have been achieved in the recent development of dihydrostreptomycin.

achieved in a reasonable period and to be lasting; (4) repeated use of the drug to be possible; and finally (5), a more questionable requirement, complete eradication of virulent infection. It would be foolish to assume that even combinations of current agents will entirely satisfy these criteria. Consequently, every new drug which appears to be suitable and safe for clinical application in tuberculosis must be submitted to the most grueling clinical-statistical assessment before it can be accepted or discarded. Otherwise, medical manpower and money may be culpably wasted to little purpose.

This problem of the assessment of efficacy of possible antituberculous agents is so pressing and so timely that most of the remainder of this lecture will be devoted to its discussion.

TECHNIQUES FOR THE ASSESSMENT OF ANTITUBERCULOUS AGENTS

Distinct advances in the laboratory methods for the testing of new antituberculosis remedies have been made over the methods of ten years ago. For *in vitro* assays we have improved media such as those of Youmans (25, 89, 90) and of Dubos and Davis (91), though it should be noted that the constituents of the latter medium may modify the chemotherapeutic action of some agents (92 to 94). For animal work we have the classical procedures of Feldman and Hinshaw (88) in guinea pigs and the introduction of mice for intravenous, intraperitoneal and inhalation infection (42, 73, 95 to 97).

In our techniques for assessing the value of new remedies in man, however, we are just emerging from the jungle of empiricism. Only a few months of enterprise sufficed to introduce tuberculin and gold into the treatment of tuberculosis on a world scale, despite inadequate early clinical trials. Nevertheless it took many years of scientific persistence to discredit these forms of therapy (12). It should be mentioned that untreated controls were rarely used for comparison in the early days of gold therapy and when they were used (particularly in one outstanding American trial (98)), the results were discouraging.

In spite of these and similar experiences, it is doubtful whether we have yet fully learned the lesson of how easy it is to ascribe to a new drug the improvement noted in a hotchpotch of tuberculous infections, without due regard to the possibility of remission from other causes. Even now, in the recent and current trials of the sulfones, streptomycin, PAS and other agents, can it be claimed that the selection of material, uniformity of treatment, and the basis of comparison for assessment of results, have always reached scientifically desirable levels? Moreover, now that experience with streptomycin has proved that drugs *can* influence human tuberculosis, there is a danger that the dearly won and healthy attitude of caution may be discarded and over-optimism and a too easy acceptance of newer measures take its place.

These considerations bring us to a key question in clinical trials, namely, under what circumstances is it necessary that a chance-selected, adequately matched group of untreated cases be studied concurrently with the cases receiving the particular treatment under investigation? If a drug prolongs life and produces apparent cure in a proportion of tuberculous infections of a type usually

associated with a one hundred per cent mortality, it is clearly evident that controls are unnecessary for evaluation of the therapy. Such a situation is provided by tuberculous meningitis and it is an interesting corollary that it is just in this and other acute fulminating forms of tuberculosis, which used to be said to constitute an unfair test of a chemotherapeutic agent, that the effect of streptomycin has been least equivocal. Untreated controls are also unnecessary where the course of the disease after administration of a drug is found to be in sharp conflict with previous clinical experience. In instances of this type there is such a prompt, rapid and easily observed reversal of the course of the infection that no doubt exists that a new factor is operating. Examples of this type of unequivocal therapeutic response are afforded by the early penicillin tests in acute pyogenic infections, and the course of tracheobronchial ulcerations or superficial sinuses after treatment with streptomycin. In the subacute and chronic tuberculous conditions, however, where improvement may characteristically occur either spontaneously or under the conventional methods of therapy, concurrent matched controls are desirable if the bias of the observer and extrinsic factors are to be excluded. For example, PAS introduced into the pleural cavity is apparently efficacious in clearing up certain tuberculous empyemata. It is possible that the observed results may represent a true effect of PAS. It is also possible, however, that the more vigorous or frequent aspiration or lavage likely to be associated with the chemotherapy *might* be the operative factor. Consequently the question cannot be answered unless controls treated similarly by aspiration or lavage alone are also studied. Likewise the effects of bed-rest must be taken into consideration in all chemotherapeutic trials in pulmonary tuberculosis.

Where provision of concurrent control cases is not considered feasible, it is necessary to have recourse to comparisons of the patient's progress under the chemotherapeutic treatment with one or more of the following: (a) with the disease trend in the same patient during a period of observation prior to treatment (for which purpose he would have to have been in bed for, say, at least two months); (b) with his expected progress had he not been treated with the drug, this being determined by independent experts on the basis of the pretreatment trend; (c) with the progress of a series of similar, but not drug-treated, cases taken from the hospital's records. These methods of comparison have been admirably used in certain investigations (66, 68) and have demonstrated the value of streptomycin *qualitatively* in a number of conditions. It is doubtful, however, if such methods can be used *quantitatively*. In order to make more precise evaluations and to answer the all important question, whether streptomycin is of *lasting* benefit in pulmonary tuberculosis, it is essential that "concurrent controls" be included. Another reason against using previous hospital records as a standard for comparison is the fact that the character of tuberculosis may change from year to year in a community, *e.g.*, with changes in the general nutritional level.

Concurrent controls have been used with good effect in explorations of the protective value of streptomycin in chest surgery. One of the projects in the

cooperative studies of the U. S. Veterans Administration, the Army, and the Navy (66) has been organized on such a basis. Controls have also been used to some extent in a test of streptomycin in pulmonary tuberculosis at Maybury Sanatorium, Michigan (99), and are a feature of the large scale cooperative trials of this drug in pulmonary tuberculosis recently started under the auspices of the Tuberculosis Study Section of the National Institute of Health. In controlled trials of this nature one can include heterogeneous types of the disease under study, provided that the total number of cases is sufficiently large to even out differences between treated and control groups. If this is not possible, the cases must be paired, which is a troublesome procedure. An alternative approach is to restrict the study to a homogeneous type of disease. Under such circumstances the numbers need not be so large for a comparability to be achieved between the treated and untreated groups, although the results will be more limited. Other conventional treatment can be added to both groups, or withheld from both. The second alternative is preferable scientifically but difficult to achieve in practice.

In Great Britain, extreme scarcity of supplies of streptomycin has up to the present prevented general distribution of the drug.[5] Although this scarcity has caused grave disquiet, it has had the advantage of providing time for the design and execution of certain controlled trials of the efficacy of the drug. In 1946, the British Medical Research Council was entrusted by the Ministry of Health with all the streptomycin that was available. In January, 1947, a number of trials in tuberculosis were started under the direction of the Council's Streptomycin in Tuberculosis Trials Committee, using 50 kg. of material imported from America. Two of the first studies will now be briefly discussed solely from the standpoint of technique of assessment.

One of the investigations was concerned with pulmonary tuberculosis. It was decided to choose a narrow and fairly homogeneous category, viz., bilateral, acute progressive tuberculosis of recent development in young adults, with expected bad prognosis and lesions which were unsuitable for collapse therapy. A number of specialist units were formed to cooperate with the Medical Research Council's central scientific staff who were to coordinate the work. The task of maintaining uniformity of the type of case under study was assigned to a central selection panel. The most important decision was that one-half of the cases selected were to be given streptomycin and one-half were to be studied in parallel as non-streptomycin-treated controls (though not in the same wards). Once a case had been selected for trial, the decision as to whether it was to be streptomycin-treated or to be a control was predetermined from a list of case numbers prepared by a statistician. A sealed envelope carrying the case number outside contained the decision, which was not known in advance even by the central staff. There was a common record sheet for the trial and a high degree of uniformity of procedures was obtained. The latter was helped by meetings of the clinicians and pathologists and by periodic visits of the central staff to

[5] A limited allocation of streptomycin for routine use has been made by the Ministry of Health to the Regional Hospital Boards since this talk was given.

the local units. It was agreed that other active treatment should be adopted only if the course of the disease so changed that it became indispensable and urgent, though every patient received bed-rest and all the usual dietetic and other symptomatic therapy. The group of investigators thus submitted itself to a considerable degree of self-discipline. It should be added that, in spite of the even chance of a case being made a control, there was every inducement for clinicians to submit their patients for this scheme, since no other practicable specific treatment and no other assured source of supply of streptomycin were then available.

This trial was completed by April, 1948, after four months' treatment and a total of six months' observation in bed of each case. The results are being analysed,[6] but preliminary tabulations show a contrast between the 55 streptomycin-treated cases and the 52 non-streptomycin-treated controls. The evaluation is based on the (provisional) percentages of improvement, of deterioration and of death, at four and at six months after the start of chemotherapy. The roentgenographic assessment was by three specialists who were unassociated with the trials and ignorant of the category of the patients and who read the films independently. Although the advantage is strikingly with the streptomycin-treated group, a proportion of the controls showed improvement, thus demonstrating the possible fallacies had a control group been omitted. Furthermore, the treated group tended to do best in the first four months while such improvement as occurred in the controls *i.e.*, without streptomycin, was usually much slower, a contrast that could hardly have been appreciated without the use of a control series. It is justifiable to conclude that this controlled trial has already yielded a clear short term result, albeit on one category of pulmonary tuberculosis and that a rather uncommon one. A long term comparative follow-up of the two groups (allowing for complications due to subsequent collapse therapy or other treatment) should add further information to this study. It should be emphasized that the use of non-streptomycin-treated controls was considered to be ethically justified in this particular trial for three reasons: (*1*) it was not possible to say with certainty that streptomycin would be advantageous; (*2*) the type of case selected was usually unsuitable for alternative active treatment; (*3*) all of the small supply of streptomycin available was being used for treatment.

The other of the British early trials of streptomycin was made in children with tuberculous meningitis and the results were published in April, 1948 (70). In this study a comparison of the fate of streptomycin-treated cases, according to duration of disease when chemotherapy was started, demonstrated the great advantage of early diagnosis and the consequent importance of concentrating limited supplies of the drug on such cases. An important comparison was undertaken to decide whether the irritating effects of intrathecal treatment could be avoided in meningitis by restricting administration of streptomycin to the intramuscular route. For this purpose, alternate cases admitted to the wards were treated by the intramuscular route alone or by combined therapy (changing from the former to the latter only if it became clear that the method

[6] A full report of this investigation has now been published (100).

was failing). The results of this inquiry answered the question decisively: combined therapy is essential and intramuscular administration alone is inadequate. Such an unequivocal answer could not have been achieved unless two series of cases had been studied in parallel under fairly uniform conditions. Evaluation of an accumulation of individual clinical experiences with a variety of types of tuberculous meningitis would have taken far longer and the conclusions would have been less definite. Moreover, although the present drastic procedure involved the loss of several lives, it will have saved many more if it serves to prevent future uncontrolled experimentation on these lines. The technique of assessment here again has been of prime importance.

Summarizing the position so far reached on techniques of evaluation, it may be stated that careful objective clinical assessment, based on previous experience with similar types of cases as a standard, should, with rigid safeguards, be adequate to demonstrate whether a new drug gives either benefit or no benefit, provided its action is more than slight. On the other hand, the inclusion of a matched control series, studied concurrently in order to permit comparisons having statistical validity, would seem imperative for a precise evaluation of a new drug's possibilities, limitations, and the degree of permanence of its benefit. Likewise, such control cases are necessary in studies of the part played by the patient's own resistance to the infection when under the drug treatment or after its discontinuation and to ascertain the best combinations of chemotherapy with the usual methods of local surgery, such as pneumothorax and thoracoplasty. It is hardly necessary to add that only for certain purposes need the control series in such investigations be untreated by the drug, as was the case in the British pulmonary trials described. In many instances, as with the meningitis series, the "controls" will receive the treatment but in a contrasting manner. It should be noted that some investigators might prefer to use another word for this application of the concept of a controlled study. Examples may be mentioned of specific problems which are capable of solution by parallel comparisons more rapidly than would otherwise be the case. One of these is the question of the optimum regimen of administration of streptomycin in meningitis. For instance, were the results in a recent series described by Dubois of Brussels (101) better than the results obtained in the British trials because of the long periods of rest from streptomycin treatment of any kind? Another question is how far prophylactic intrathecal treatment may prevent the complication of meningitis in cases of acute miliary tuberculosis.

A further point not yet generally appreciated is that the problem of the techniques for assessing new drugs in tuberculosis is in some ways more difficult now that we have in streptomycin an agent that is known to be decisive in certain conditions. For example, *assuming an adequate supply of streptomycin*, it is questionable whether it is ethically justifiable to withhold this drug and to substitute an unproved one in the treatment of tuberculous meningitis. It is more justifiable to give streptomycin alone in one such series and streptomycin plus the new drug in another, although assessment may well be confused under such circumstances. On the other hand, in certain types of pulmonary tuberculosis,

it might be ethically correct to study a drug-untreated control series, at least for a time, against the treated series, in order to obtain a first evaluation of the new drug. As an alternative system the new drug could be tested in comparison with streptomycin. Unquestionably this whole problem of investigation techniques will itself develop and change as new remedies grow in number and variety.

LOOKING AHEAD

I have tried in this talk to take stock of the status of antituberculosis chemotherapeutics in a general way and to discuss the technical equipment for the clinical assessment of individual drugs. The future needs of chemotherapy will be conditioned by the changing pattern of tuberculosis. There is no doubt that a prime objective will be a drug, a combination of drugs, or a technique of administration of drugs, for use in minimal pulmonary tuberculosis. In order for such a treatment to be satifactory, it must produce results rapidly, safely, and without appreciable risk of the emergence of drug-resistant infecting strains. The prospects for the development of treatments of this kind are reasonably good. The very variety of the promising chemotherapeutic agents already under examination is a good omen for the development of better ones, whether they be found through synthesis by the organic chemist or by Nature. Perhaps a number of such drugs will come to supplement or even to replace streptomycin, though this antibiotic will retain its place of honor in medical history.

Even should our hopes of successful chemotherapy be fulfilled, this weapon is still unlikely by itself to lead to the eradication of tuberculosis. For tuberculosis tends to be a chronic infection firmly implanted in the tissues, and chemotherapy must be considered as only a part of the complex of treatment that has been built up through the years. The importance of the contact factor and the contribution of the patient's inborn and acquired resistance to the infection, in other words the vital function of prevention in this fight against tuberculosis, must also be always borne in mind. In a study of statistics in large towns in England, the pulmonary tuberculosis mortality in young women was found to be nearly twice as high in the most overcrowded boroughs as in the least overcrowded and doubtless a similar relationship exists in this country. Thus in the control of tuberculosis the technique of living is as important as the technique of treatment.

Finally, let us never forget to regard tuberculosis as a world problem. This disease is perhaps being defeated in some parts of the world, but, while the death rate per 100,000 is 42 in New York City and 44 in Copenhagen, it is 132 in Glasgow, 135 in Rome and 275 in Warsaw. Moreover the Warsaw rate is nearly twice what it was before the destruction due to war. In the whole world there are perhaps some ten to twenty million sufferers from active tuberculosis. There is plenty of work to do. I believe that this meeting of key men and women will provide inspiration for us all to go forward in this task of peace.

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THORACOSCOPY AND CLOSED INTRAPLEURAL PNEUMONOLYSIS[1,2]

O. C. BRANTIGAN

INTRODUCTION

Although references to pneumothorax as a therapeutic procedure date back to antiquity, it became a generally popular method of treatment for pulmonary tuberculosis about 1910 (1). Since intrapleural adhesions often interfere with adequate pneumothorax collapse of the lung, it is natural that a method of freeing the lung from the effects of adhesions would be developed. Friedrich (2) probably did the first pneumonolysis in 1908. It was the open type of pneumonolysis. The closed method of releasing adhesions was developed by Jacobaeus and Beiter (3) in 1913 and, interestingly enough, the procedure had to prove its superiority over the open type of operation. In the beginning there was no real difference in the indications for the two operations. The closed method of intrapleural pneumonolysis became the accepted procedure in Europe before it did in the United States but by 1929 Matson (4) had established it in this country as the operation of choice for freeing intrapleural adhesions. By 1934 the open method of pneumonolysis had been rather generally discarded because the morbidity and the mortality were extremely high after the operation. In spite of the high mortality and morbidity, Anderson and Alexander (5), Dohnanyi (6), and Hoppe (7) maintained that the open method procedure, when properly used, had its place in the treatment of pulmonary tuberculosis. In 1946 the author (8) reported his experience with 15 patients who were subjected to the open type of pneumonolysis. Since that report the open method has been used on 8 additional patients. There have been no serious immediate or late postoperative complications and the end results have been gratifying. The introduction of new chemotherapeutic agents makes the operation even more desirable and probably will extend its use to include patients who have more extensive adhesions. It seems, however, that the other rigid indications should be maintained. The open type of operation should not be considered unless there is failure to free the lung by the closed method.

Since the work of Matson (4), the closed method of intrapleural pneumonolysis has been accepted without controversy. It has been ably supported by Moore (9), Goorwitch (10), Bayliss (11, 12), Kunstler (13), and others. There has, however, been considerable discussion concerning the efficiency of the one or two piece pneumonolysis instrument. There has also been much controversy as to the advantages or disadvantages of galvanocautery compared with high frequency electrosurgical cutting current (electrosurgery).

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[2] Presented before the Medical Section, as part of the symposium on Surgery—Trends—Late Results, at the 44th Annual Meeting of the National Tuberculosis Association, New York, New York, June 16, 1948.

OPERATIVE TECHNIQUE

It makes little difference whether there are one, two, or more trocar holes through the chest wall. The size of the trocar hole is unimportant, particularly when compared with an open incision into the pleural cavity. At the beginning of the operation the use of two cannulas permits the inspection of the pleural cavity from two different points of view. The adhesions may appear simple or impossible of release, depending upon the angle from which they are viewed. The two piece instrument eliminates the fixed position of the cutting instrument in the field of vision. Thus, a close-up view of the actual cutting can be obtained or in a moment the whole field of operation can be examined without interrupting the operation. After using both instruments, the two piece type is now employed exclusively.

Galvanocautery, or the electrosurgical cutting current, can be used as the cutting method with either the one or two piece pneumonolysis instrument. It is usually claimed that tissue destruction is greater with galvanocautery and that the separation of areas of necrosis may lead to secondary hemorrhage or spontaneous pneumothorax. Actually, the area of tissue destruction with either galvanocautery or electrosurgery depends upon the intensity of the heat in the former and the strength of the current in the latter. The extent of tissue destruction, therefore, depends more upon the individual operator than it does upon the method employed. Heat and smoke produced by galvanocautery properly employed are insignificant. The time consumed in completing an operation by either method depends upon the patient and the operator. Should brisk hemorrhage occur from a blood vessel that is less than one of the great vessels, it can be controlled more easily with the electrosurgical method. If hemorrhage can be controlled with electrosurgery, however, it also can be controlled with galvanocautery. In the event a great vessel is opened, neither method will control the hemorrhage. There is no electrical shock or muscle spasm from galvanocautery, and undoubtedly there will be none with electrosurgery if the apparatus is handled correctly. Since the galvanocautery is the simpler apparatus, mechanical failure probably will occur less frequently than with the electrosurgical unit. (After using both types of cutting instruments the galvanocautery is now used exclusively.) When operating with the galvanocautery equipment, an electrosurgical electrode could be made available for use in case of bleeding. It seems quite likely that an electrosurgical electrode would always be on hand if bleeding actually were troublesome when using galvanocautery.

The heat of the cautery or the strength of the electrosurgical cutting current must be regulated according to many factors since in using either method the tissues may be cut so quickly that bleeding occurs or so slowly that excessive charring results. In general, fibrous tissue, fatty tissue, and edematous tissue require a hotter cautery or a stronger cutting current. When holding the cutting instrument constantly in contact with the tissue or when attempting to keep a greater area of cutting surface against the tissue to be cut, a hotter cautery or a stronger cutting current is required. Should bleeding occur, it can be controlled easily by the electrosurgical method; if cautery is being used, however, it is often controlled most readily by reducing the heat below that used in operating. The cold electrode is placed firmly against the bleeding point and then the current is turned on. The cautery point is held in place until the tissue is charred. As the heat is reduced, it will not cut deeper into the tissue. If it cut deeper into the tissue, it might cause additional bleeding. The charred tissue will adhere to the cautery point. Therefore, when the point is to be removed, it must be done gently and with the heat of the cautery point increased. Bleeding often can be controlled adequately by the injection of a generous amount of procaine with epinephrine solution at the site of bleeding. The author has never observed bleeding from the lung side of a released adhesion.

All surgery that is accomplished though an eyepiece for vision remains concealed from onlookers or critics. The closed type of pneumonolysis is no exception and, as sterility must be carefully guarded, it is difficult, if not impossible, to photograph the operation. The actual operative technique, perhaps, will depend upon the individual operator. Nevertheless, certain aspects of the operative procedure should be discussed. After a patient who had undergone a closed pneumonolysis developed a pure *Staphylococcus albus* empyema, sterile hoodlike masks have been employed (figure 1). The operator wears glasses sterilized by soaking in alcohol (figure 1). The operation is always begun with the patient in the lateral position. A wide area of skin is prepared. Well within the cleaned skin area, procaine wheals are made in which towels can be anchored to the skin by towel clips. The towels are then placed so as to give access to an area of skin anteri-

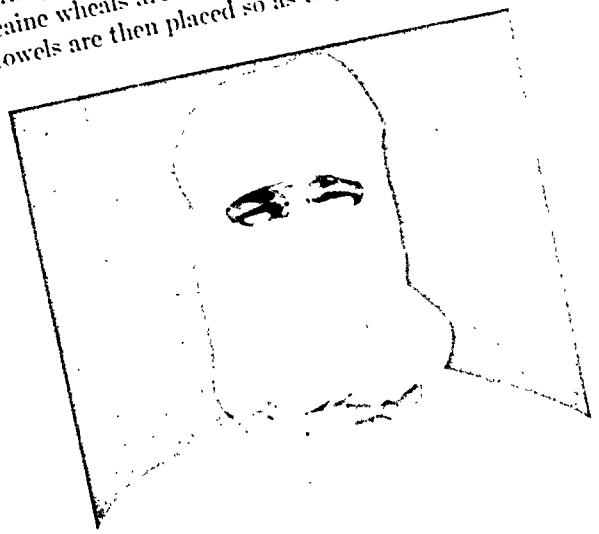


FIG. 1 The photograph shows the operator dressed for closed pneumonolysis. He is wearing a sterile hood and sterile eyeglasses.

orly, laterally, and posteriorly. The towels are anchored to the skin so that they cannot shift as the patient is turned in various positions (figures 2 and 3). The first trocar and cannula puncture site is selected in about the third interspace at approximately the anterior axillary line (figures 4 and 5). Roentgenograms suggesting an adhesion in the region may cause a variation of the first puncture site. The second trocar and cannula puncture site is selected after adhesions have been thoroughly examined, usually it is inferior and posterior to the first cannula, just anterior to the inferior angle of the scapula. Care is exercised so that the second site selected is not too close to existing adhesions. It has been found that a cannula inserted posteriorly is to the inferior angle of the scapula and accordingly is avoided except for [illegible] adhesions. The two cannulas [illegible] are also selected so that [illegible] posteriorly [illegible] the operator [illegible] lateral, and posterior [illegible]

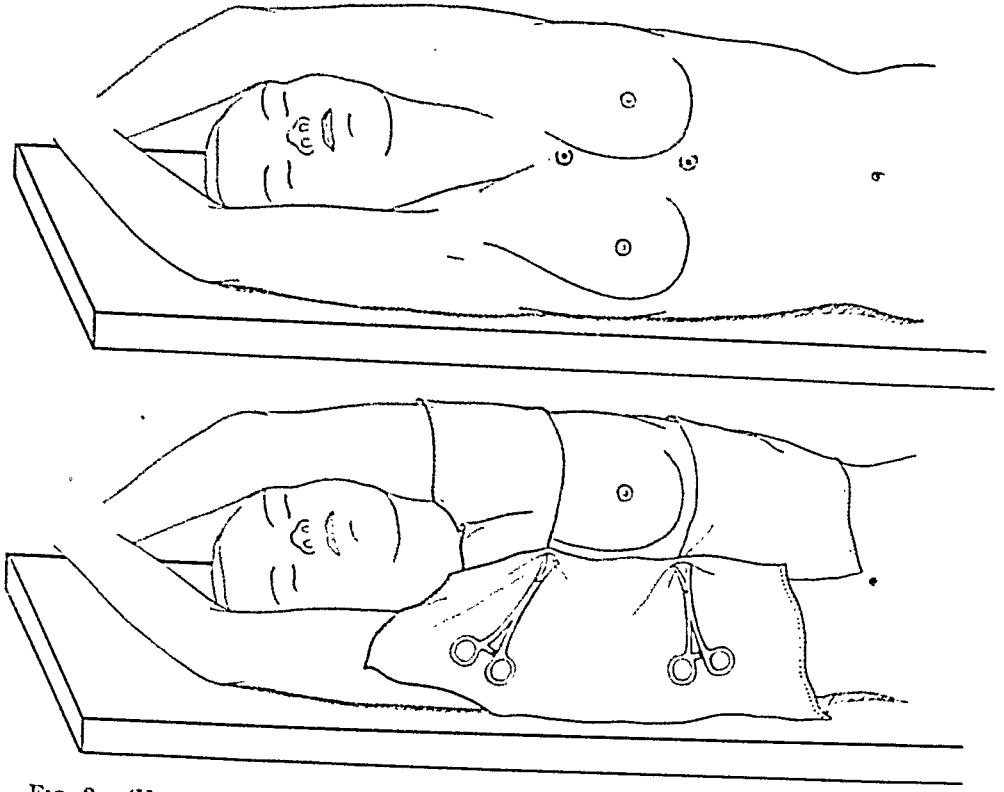


FIG. 2a. (Upper) An anterior view of the large area of skin prepared for operation. The procaine wheals may be seen.

b. (Lower) The method of draping the patient for operation is illustrated. The towels are anchored to the skin by towel clips inserted into the skin at the site of the procaine wheals.

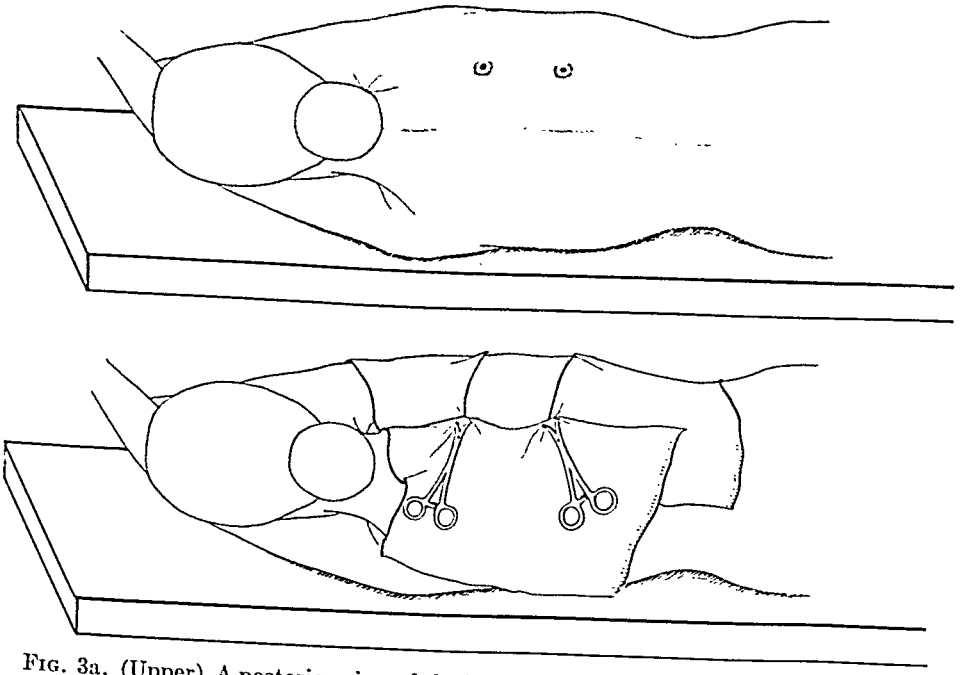
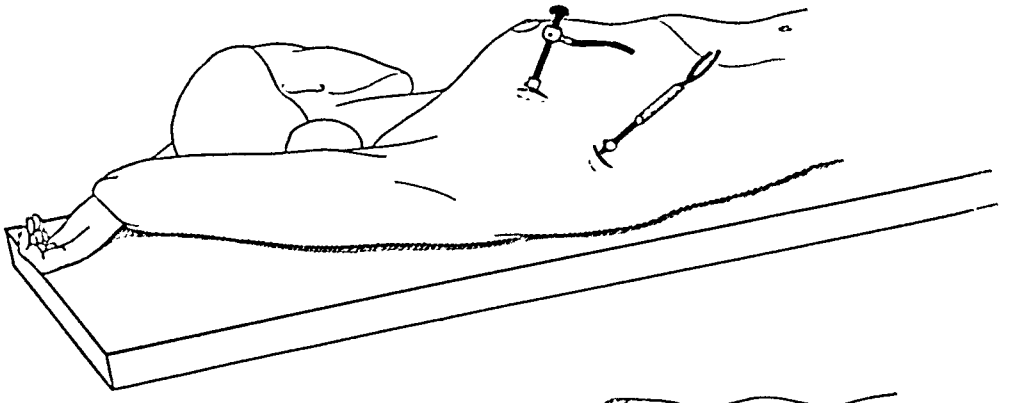
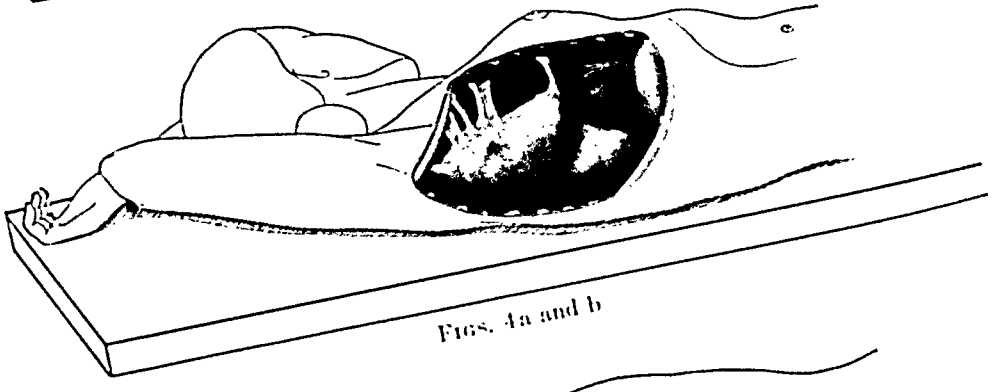


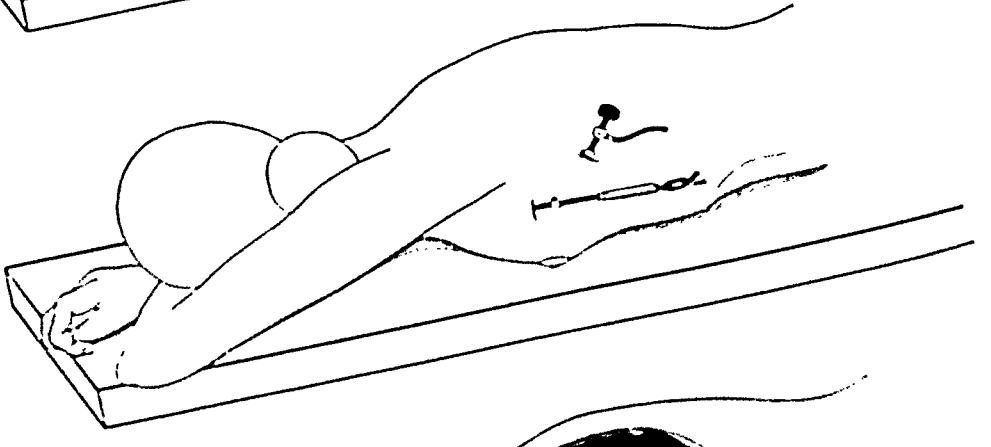
FIG. 3a. (Upper) A posterior view of the large area of skin prepared for operation. The procaine wheals may be seen.

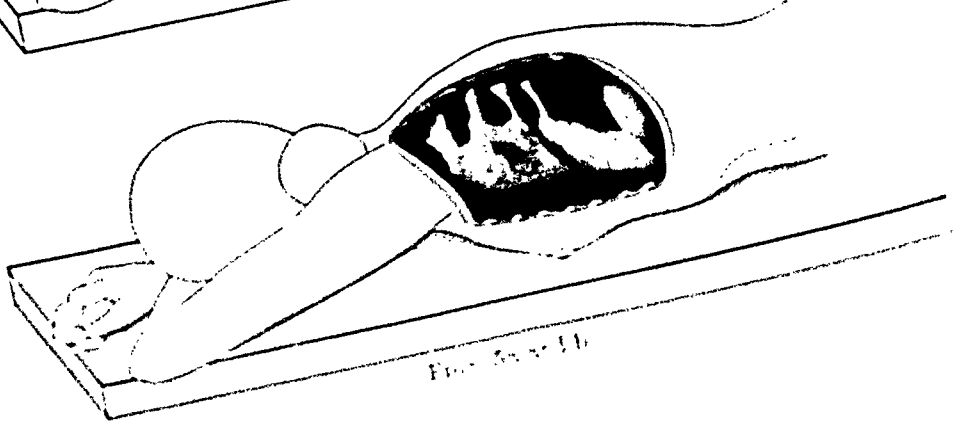
b. (Lower) The method of draping the patient for operation is illustrated. The towels are anchored to the skin by towel clips inserted into the skin at the site of the procaine wheals.





FIGS. 4a and b





FIGS. 5a and b

As in the presence of a pneumothorax the lung falls by gravity, it is important to put the patient in a position that will cause the weight of the lung to pull upon or stretch the adhesion under surgical attack (figures 4 and 5). Therefore, the patient must be prepared and draped and the cannulas inserted in such a manner that he can be placed in any and all positions; head up, head down, prone, supine, lateral, and even at times with the patient so close to the edge of the operating table that the contralateral side of the chest is uppermost.

If the weight of the lung is allowed to pull upon or stretch the adhesion under surgical attack it can be released from the chest wall more easily and safely. As a portion of the adhesion is freed, it will fall away from the chest wall and permit better access to the remaining portion of the adhesion (figure 6). Bayliss (12) recommends insertion of a third cannula in order that traction can be made upon the adhesion under surgical attack. The writer has employed this method when for some other reason a third cannula has been necessary. The application of traction upon the adhesion under surgical attack, as Bayliss so adequately describes, is a most effective method and perhaps should be adopted as a routine procedure in closed intrapleural pneumonolysis.

Adhesions are removed from the chest wall as described by Carter (14); they should not be cut or divided. Even the simple adhesions are attacked in this manner for it keeps the operator in practice. The parietal pleura and endothoracic fascia are divided near the base of the adhesion and the adhesion is then released in the extra-endothoracic plane. This usually is an areolar tissue plane. The fascia covering the intercostal muscle can be recognized on the chest wall side (figure 6). In patients where pneumothorax has been of short duration, the plane is identified easily. The areolar tissue melts like butter ahead of the cautery and rarely is bleeding encountered. Small capillaries must be present since *cold stripping* causes enough bleeding to obscure the fine identification necessary to maintain the plane. Dissection in the extra-endothoracic fascial plane of areolar tissue with hot cautery has been so successful that the author completely gave up the cold stripping method (15, 16). In order to release adhesions in the extra-endothoracic fascial plane, it is necessary to have the adhesions on a stretch. This method of release of the adhesions is safe as one cannot injure the lung even though a portion of it is adherent to the chest wall. The intercostal vessels are separated from the plane by the internal intercostal muscle except posteriorly, where the posterior intercostal membrane replaces

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FIG. 4. The drawings illustrate the patient placed in the supine position on the operating table. The patient is placed close to the edge of the table.

a. (Upper) The pneumonolysis instruments are placed in a position that will allow manipulation in the supine, lateral, or prone position. The eyepiece and cautery can be used interchangeably in either cannula.

b. (Lower) The drawing shows that the lung falls by gravity to the posterior part of the chest. Adhesions from the anterior aspect of the lung to the anterior chest wall are placed on tension by the pull of the lung. An adhesion on tension is released more easily by closed pneumonolysis. If adhesions are present from the posterior aspect of the lung to the posterior chest wall they cannot be seen and hence could not be released.

FIG. 5. The patient is placed in the prone position on the operating table, close to the edge of the table. Sometimes the torso is extended over the edge of the table, thus permitting freer manipulation of the pneumonolysis instruments.

a. (Upper) The pneumonolysis instruments are in place as indicated in figure 4a.

b. (Lower) The drawing indicates the lung falling anteriorly by gravity. The posterior adhesions are placed on tension by the weight of the lung. Anterior adhesions cannot be seen and hence could not be released in this position.

the internal intercostal muscle. At this posterior location and anteriorly at the internal mammary vessels, both the veins and arteries are subpleural and vulnerable. In these locations extrapleural or extra-endothoracic fascial separation must be abandoned and the adhesion divided. When the adhesions are attached to the large blood vessels or to the trachea or esophagus, they must be divided, since release in the extrapleural or extra-endothoracic plane is dangerous if not impossible. The partial release or partial division of an individual adhesion is avoided when possible. On occasion, however, it is necessary

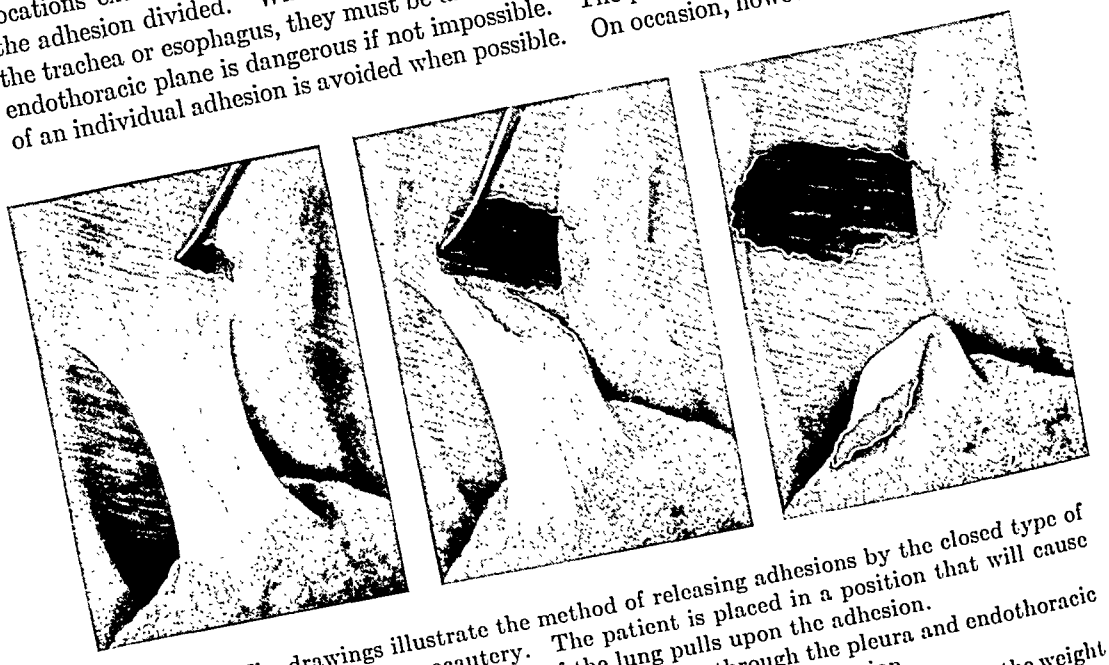

FIG. 6. The drawings illustrate the method of releasing adhesions by the closed type of pneumonolysis using galvanocautery. The patient is placed in a position that will cause tension on the adhesion. The weight of the lung pulls upon the adhesion.

a. (Left) The adhesion is first attacked by cutting through the pleura and endothoracic fascia approximately a centimeter away from the base of the adhesion.

b. (Centre) As the adhesion is released in the extra-endothoracic fascial plane the weight of the lung pulls the released portion of the adhesion away from the chest wall, allowing free access to the remaining portion of the adhesion. The extra-endothoracic fascial plane is areolar tissue, which melts like butter ahead of the hot cautery. In this plane bleeding does not occur unless the lung is stripped by the cold cautery. The internal intercostal muscle or the intercostal membrane usually can be identified without difficulty. If the adhesion is divided in its smallest part, injury to the lung parenchyma will occur if the lung parenchyma is drawn into the adhesion. When the adhesion is released in the extra-endothoracic fascial cleavage plane, the lung parenchyma cannot be injured even if the visceral and parietal pleura are intimately adherent.

c. (Right) The area of chest wall denuded is generally several times greater than the smallest part of the released adhesion.

to begin the release or division before the operator can be certain about his ability to completely release or divide the adhesion.

The objective of the operation is to free the lung completely from the chest wall, mediastinum, and even from the diaphragm. At one time it was thought that the duration of the operation was important but such reasoning has been discarded. The operation requires from twenty minutes to two and one-half hours. All adhesions are released if possible. There is never a deliberate plan to attempt to complete the freeing of the lung at a subsequent operation. Subsequent operations have been rather disappointing.

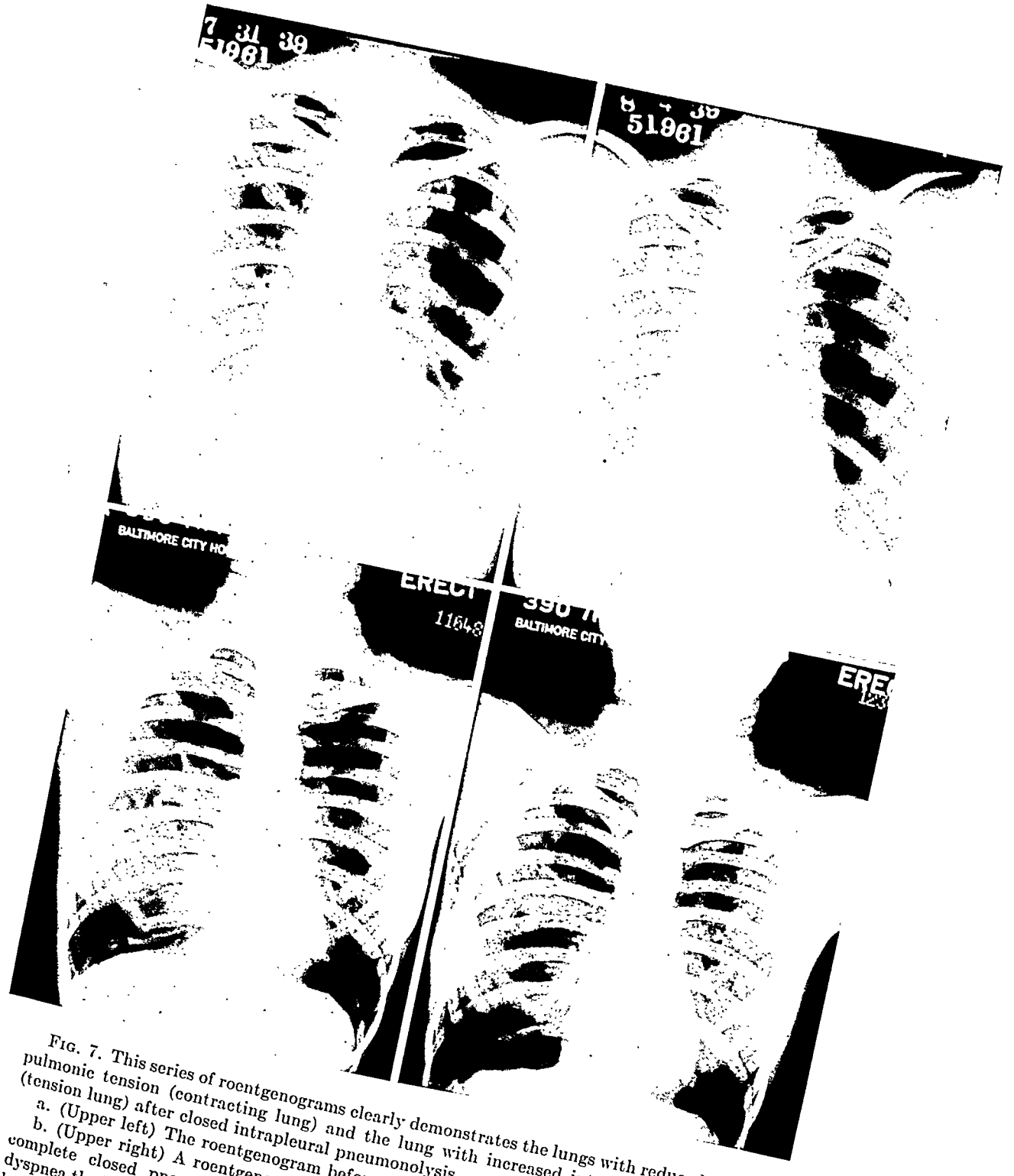

FIG. 7. This series of roentgenograms clearly demonstrates the lungs with reduced intrapulmonic tension (contracting lung) and the lung with increased intrapulmonic tension (tension lung) after closed intrapleural pneumonolysis.

a. (Upper left) The roentgenogram before thoracoscopy was done.

b. (Upper right) A roentgenogram of the same patient about twenty-four hours after complete closed pneumonolysis. There is no subcutaneous emphysema. Because of dyspnea the intrapleural pressure was measured. On inspiration the pressure was found to be −28 cm. of water. As indicated, the lung is atelectatic.

c. (Lower left) The roentgenogram before thoracoscopy was done.

d. (Lower right) The same patient about twenty-four hours after complete pneumonolysis. The subcutaneous emphysema and the tendency of the lung to re-expand can be seen. Subcutaneous emphysema indicates increased intrapleural pressure.

When the operation seems completed the patient is routinely turned successively in the prone, lateral, and supine positions for final inspection of the lung and pleural cavity. Care is exercised not to overlook an adhesion between the two cannulas. The instruments are removed. A silk suture is used to close only the skin at the puncture site. If the lung has been freed completely, 200 to 300 cc. of air are introduced into the pleural cavity and a small dressing is applied. Pressure dressings have not been employed since the cause of subcutaneous emphysema has been understood (17, 18).

The patient is returned to his bed and encouraged to move and turn often and to sit up in bed. This is carried out with great care in order to avoid having the lung lie against the chest wall in one position long enough to become adherent. At operation the lung is judged to have increased, normal, or decreased intrapulmonic pressure (figure 7). A lung with increased intrapulmonic pressure will inflate and cause increased intrapleural pressure, thus forcing the intrapleural air into the tissues through the puncture sites (figures 7c and 7d). If at operation the lung is judged to be of the increased tension type or if there is a question of increased intrapulmonic tension, the patient is given 0.5 cc. of epinephrine 1:500 in oil hypodermically every four hours. Epinephrine is effective as bronchial spasm is the most frequent cause of the increased tension type of lung. Moreover, bronchial spasm may also play a part even though organic bronchial disease may be the exciting cause. Intrapleural operations as well as operations upon the lung frequently initiate temporary bronchial spasm. The patient is examined a few hours after operation and, if the lung type has been judged incorrectly, there may be moderate subcutaneous emphysema, for which epinephrine in oil is given. Most patients have a slight degree of subcutaneous emphysema because moving, coughing, talking, vomiting, defecation, and other actions cause increased intrapulmonic pressure with a corresponding elevation of intrapleural pressures and the escape of air into the tissues through the puncture sites. The state of lung collapse is usually checked by fluoroscopic examination the day after operation. It is unusual for the patient subjected to the closed type of pneumonolysis to require an air injection until several days after operation.

On rare occasions a lung completely freed of adhesions but not markedly collapsed will adhere to the parietal pleura in a few hours. The adherence will be so tight in about twenty-four hours that it cannot be freed by increased refills of air or a change in position. At operation it is difficult to detect the patient whose lung will react in this manner. It seems that in the rare patient even though bleeding is not the cause it is impossible to keep good vision through the eyepiece, since a film forms over the intrapleural lens when it touches any tissue within the pleural cavity. This film cannot be cleared from the lens by brushing it against the lung, as is the usual practice. Such difficulty in keeping the lens clear probably indicates an unusual stickiness of the body fluids and may account for the quick adherence of the lung to the parietal pleura. This is perhaps the opposite of a pleura that reacts by a thin serous exudation. When the sticky type of pleural cavity is found and easy adherence of the lung to the parietal pleura is predicted, steps are taken to avoid the mishap. The lung is collapsed to a greater degree than usual by air injection into the pleural cavity at the completion of operation and the patient is ordered to change position every two hours, day and night, for the first twenty-four hours after operation. Since recognizing the possibility of the sticky type of pleura and lung and adopting the above method of postoperative treatment, in no patient has a lung become adherent to the parietal pleura after pneumonolysis.

Indications for Pneumonolysis

There is only one indication for closed intrapleural pneumonolysis: the patient must be a candidate for artificial pneumothorax, and intrapleural adhesions are

preventing adequate collapse of the lung. It is not within the scope of this paper to discuss the indications for artificial pneumothorax. It is important, however, for the surgeon who performs pneumonolyses to recognize certain contraindications for artificial pneumothorax, which is another way of stating the contraindications for pneumonolysis. It is becoming generally realized that artificial pneumothorax should not be continued if after its initiation one of the following develops: large tension cavity (figure 8 a); an opaque lung (figure 8 b);

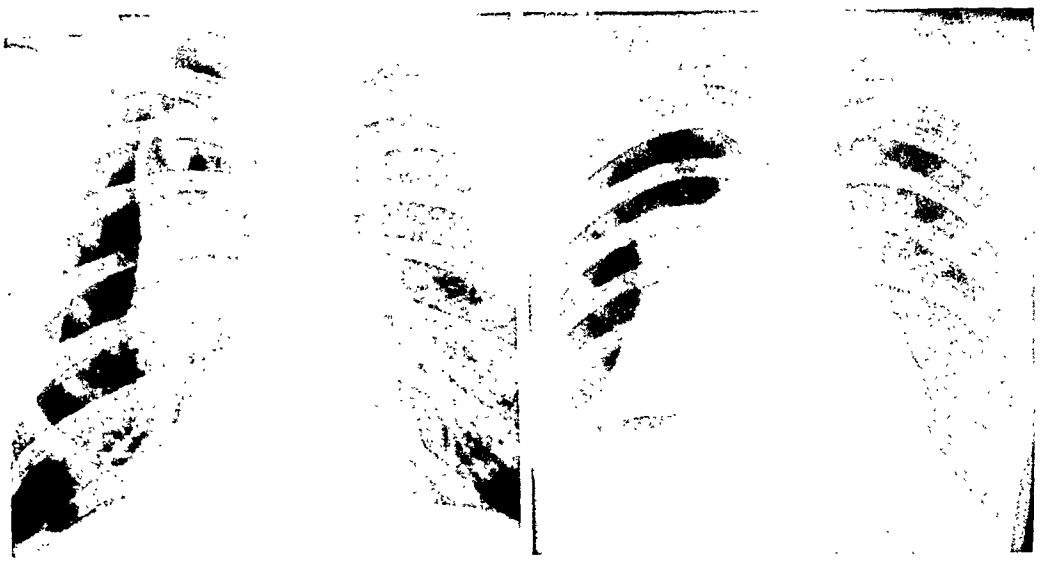

FIG. 8. Two entirely different types of pulmonary tuberculosis are illustrated. In both types of the disease closed intrapleural pneumonolysis as well as pneumothorax are contraindicated.

a. (Left) The roentgenogram reveals a large tension cavity. Under protest of the surgeon the patient was subjected to closed intrapleural pneumonolysis. The lung was freed completely. After bronchoscopic aspiration the cavity appeared to close. The lung became atelectatic, intrapleural fluid developed, which was followed by empyema, a bronchopleural fistula and death. This sequence of events can be expected in patients with disease of this type.

b. (Right) The atelectatic or solid lung, especially with the open cavity, can be expected to follow the clinical course outlined in *a*. Pneumonolysis was refused this patient. Pneumonectomy was accomplished with a successful outcome.

empyema; or the rapid formation of a pleural effusion with or without fever. If the patient is a candidate for artificial pneumothorax and adhesions are found between the lung and the parietal pleura, he should be offered the advantage of closed pneumonolysis. There are no innocent adhesions. A pneumothorax should not be abandoned because of adhesions until a thoracoscopy has been made to determine whether or not the adhesions can be released. Whether adhesions can be released or not cannot be determined by roentgenograms or fluoroscopy (figure 9).

Progress has been slow in developing the routine of early thoracoscopy or pneumonolysis after the initiation of pneumothorax. In 1939 the usual time

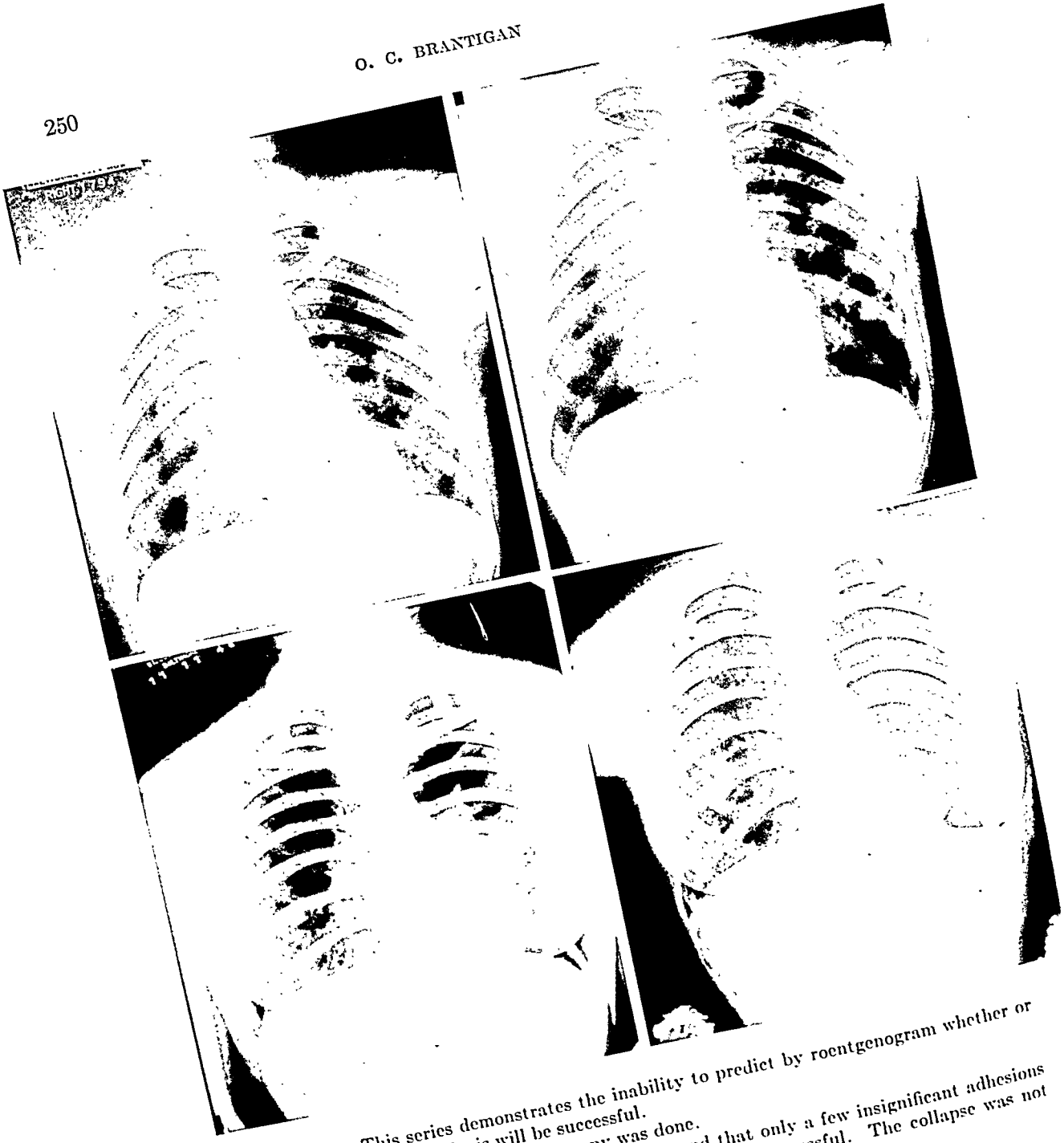

FIG. 9. This series demonstrates the inability to predict by roentgenogram whether or not closed pneumonolysis will be successful.

a. (Upper left) Before thoracoscopy was done.

b. (Upper right) At thoracoscopy it was found that only a few insignificant adhesions could be released. Closed pneumonolysis was thus unsuccessful. The collapse was not changed materially.

c. (Lower left) Before thoracoscopy was done.

d. (Lower right) A complete pneumonolysis was done. The collapse was satisfactory after operation.

interval from the introduction of pneumothorax to closed pneumonolysis was three to six months (1, 19). In patients who have an ineffective collapse of the diseased lung, it seems unwise to permit such a long delay in bringing the disease under control. The time interval has gradually been made shorter. The only requirement at present is that there be enough pneumothorax space in which to manipulate the instruments. Patients have been accepted for pneumonolysis within three days of the initiation of the pneumothorax. It was feared in the beginning that re-expansion or the loss of pneumothorax space would occur more frequently, but it does not. It is evident that more patients can have their lung freed completely from the parietal and mediastinal pleura if the pneumonolysis is done within two weeks of the initiation of pneumothorax. In that period, adhesions and endothoracic fascia do not have a chance to thicken or undergo fibrosis. When early pneumonolysis is accepted and practiced, a patient can have a pneumothorax induced, a closed or open type of pneumonolysis, and the diseased lung under control in two to three weeks. In addition to the great advantage to the operator, the time saved the patient would justify the adoption of early pneumonolysis.

RESULTS

The present series concerns 608 patients operated upon between October, 1938 and December, 1946, or an eight year period. The follow-up period on these patients extends to December, 1947. The patients have been obtained from all the sanatoriums in Maryland, both privately owned and state and city controlled, as well as from doctors in the private practice of medicine in Maryland.

The report is marred by the inability to make the study complete. It has been impossible to find the charts of 97 patients. Unfortunately, many of the lost charts overlap the 109 patients reported in 1941 (20). The charts are missing for a few patients operated upon in 1938. The source of the patients and the place of operation have been highly diversified. The method of record keeping and changes in systems of filing have accounted for the high percentage of missing charts. The analysis of operative data thus concerns 511 patients. Certain facts now desired were not recorded in the notes of operation during the early years. Therefore, in 45 cases it is unknown whether the operation was thoracoscopy only or pneumonolysis, complete or incomplete. For the same reason the condition of the pleura at the time of operation is not known in all patients.

It seems logical that the patients be divided into Negroes and whites and that the patients from sanatoriums privately owned and from doctors in private practice be separated from those patients from sanatoriums under state or city control. All the patients have been operated upon by the writer. All but 3 were operated upon under local anesthesia. In tables 1, 2 and 3 the results are given as briefly as possible. In table 1 the years of follow-up are indicated since they extend to December, 1947. There were 301 females and 210 males. The youngest patient was five years of age and there were 137 patients twenty years of age or younger. The oldest patient was fifty-eight years of age and only 9

TABLE 1
Composition of series

Source of patients	Year of operation	Number of patients	Operation: Pneumonolysis: Complete	Operation: Pneumonolysis: Incomplete	Operation: Thoracoscopy only	Living and well pneumothorax maintained: Yes	Living and well pneumothorax maintained: No	Living and sick pneumothorax maintained: Yes	Living and sick pneumothorax maintained: No	Condition unknown	Dead
Private doctors and sanatoriums	1939 through 1941	20	11	6	3	3 1 1	3 1 1	2 0 1	2 1 0	1 3 0	0 0 0
	1942 through 1944	94	48	33	13	19 5 2	20 11 4	1 3 1	2 2 0	5 4 2	1 8 4
	1945 through 1946	56	23	21	12	13 8 2	3 3 3	2 3 1	0 2 2	3 5 4	2 0 0
State and City Sanatoriums	1939 through 1941	60	33	17	10	2 1 0	2 6 4	1 0 0	0 1 0	26 4 5	2 5 1
	1942 through 1944	76	37	27	12	10 3 0	12 6 2	1 1 0	1 5 2	7 6 6	6 6 2
	1945 through 1946	27	14	8	5	6 4 2	1 1 1	0 0 0	1 0 1	6 3 1	0 0 0
Negro	1939 through 1941	125	60	41	24	1 0 1	14 9 3	0 1 0	3 1 1	7 6 2	35 24 17
	1942 through 1944	8	5	1	2	0 0 0	0 0 0	0 0 0	0 0 1	3 0 1	2 1 0
	1945 through 1946	0									

were fifty years old or older. The present status of 104 patients is unknown. One hundred thirty-six of the patients are dead (table 2). Of this group 131 died of tuberculosis in some form and 5 died of causes not related to tuberculosis. There are living and sick 45 patients and 29 of these are ill because of disease in the contralateral lung. One hundred ninety-eight patients are living and well. In the living and well group 84 patients still have a pneumothorax and 114 do not

TABLE 2

Late complications and cause of death

OPERATION	NUMBER	EMPYEMA						BRONCHO-PLEURAL FISTULA		OBLITERATIVE PLEURITIS		COMPLICATIONS NOT PLEURAL		DEATH	
		Tuberculous		Mixed		Pyogenic								Tuberculosis	Other
		L	D	L	D	L	D	L	D	L	D	L	D		
Complete	231	2	7			1	1			1		4	7	44	4
Incomplete	150	8	5		1			1	1	2		8	7	43	1
Thoracoscopy	85	4	1		1			2	1			1	7	24	
Unknown type	45				1							3	1	20	
Total	511	14	13		3	1	1	3	2	3		16	22	131	5

TABLE 3

Time from initiation of pneumothorax to closed pneumonolysis

DAYS	NUMBER OF PATIENTS	THORACOSCOPY ONLY	COMPLETE PNEUMONOLYSIS	INCOMPLETE PNEUMONOLYSIS	UNKNOWN
0 to 5	8	2	3	3	0
6 to 10	34	1	16	17	0
11 to 20	55	6	29	20	0
21 to 40	53	8	15	17	13
41 to 60	82	4	33	17	28
60 plus	155	34	82	39	0
Unknown	124	30	53	37	4
Total	511	85	231	150	45

have a pneumothorax. Of the 114 all have had their disease arrested by pneumothorax, except 6 patients who underwent major surgery in order to arrest their disease. In this group the supplemental phrenic operations have not been tabulated. Approximately two-thirds of the pneumothoraces in the entire group, including those now well and sick were abandoned as an elective procedure. Two pneumonolyses were done on 61 patients and the second operation resulted in complete pneumonolysis in 21 of the patients. Three pneumonolyses were done on 8 patients and complete pneumonolyses were accomplished at the third operation on 3 of the patients. The early complications were rather few. Hemorrhage of mild degree occurred in 28 patients. It is estimated that in the most

severe hemorrhage not more than 100 cc. of blood were lost. In the majority only a few cubic centimeters of blood were lost. One patient had a spontaneous leak of air from the lung about twelve hours after operation. The lung was made to completely re-expand by catheter suction. The loss of pneumothorax space was the only ill effect, even though the patient had a bilateral pneumothorax at the time of the accident. The pneumothorax space was lost in 3 other patients. One patient died three days after thoracoscopy without pneumonolysis. The cause of death was never accurately determined. One patient developed pyogenic empyema and subsequently lost her life. In 6 patients the lung was re-attached to the parietal pleura in the first twenty-four hours after complete pneumonolysis. Persistent pleural fluid (not empyema) was found in 28 patients. Tuberculous empyema (Table 2) occurred in 27 patients and 13 of these died. Where the pleura was delicate and normal at the time of operation, tuberculous empyema occurred in 1.6 per cent of the cases. Where the pleura was abnormal, that is, inflamed or presented tubercles, fibrin or fluid, the incidence of tuberculous empyema was 7 per cent. Spontaneous pneumothorax occurred in 6 patients. Five of these developed bronchopleural fistulas, 3 developed one suffered no ill effects. Of the 5 with bronchopleural fistulas, whereas mixed infection empyema but are alive and the other 2 patients are dead. The development of bronchopleural fistulas with mixed infection empyema or death occurred long after operation and may have been unrelated to operation. A pleurocutaneous fistula was found in one patient. Obliterative pleuritis occurred in 3 patients, with a late loss of pneumothorax space. Other complications did not involve the pleura and included such conditions as hemoptysis and tuberculous meningitis.

It would appear that whether or not the patient is cured of pulmonary tuberculosis depends upon the effectiveness of pneumothorax as a method of treatment and therefore late therapeutic results might not be considered relevant in a discussion of the closed type of pneumonolysis. There are many factors closely associated with this closed operation, however, that will affect the end result. The various early and late complications and whether the lung was partially or completely freed are factors directly associated with the operation. The patient who is not a candidate for artificial pneumothorax should not have a pneumonolysis. Moreover, it is possible to have successful pneumonolysis and artificial pneumothorax on one side and yet have the patient die from disease in the other lung. Furthermore, disease in the other lung might necessitate the abandonment of an effective pneumothorax following pneumonolysis. An effective pneumothorax after pneumonolysis could be handled badly and produce a poor result. In spite of all these factors it seems proper that the closed intrapleural pneumonolysis be held chiefly responsible for the final results obtained. It would be extremely interesting to compare a group of patients who have effective pneumothorax without pneumonolysis with a group of patients subjected to pneumonolysis which rendered the pneumothorax effective.

It is obvious that there is no satisfactory classification for tuberculosis and, therefore, there is no adequate means for comparing groups of patients. The

Negro group of patients show an overall death rate of 67.5 per cent as compared to the 15.1 per cent (table 1) for white patients. However, the group of white patients under the care of private doctors and private sanatoriums had an overall death rate of 10.5 per cent compared to 22.2 per cent (table 1) for the state and city sanatoriums group. It is perhaps impossible to correlate these series-mortality figures with the observations of other investigators. It may be desirable, however, to present the mortality rate for pulmonary tuberculosis in the state of Maryland. The fate of all patients discharged from the state sanatoriums was followed for five years beginning in 1934 (21). After five years the mortality rate in the white patients was as follows: far advanced disease, 62 per cent; moderately advanced disease, 25 per cent; and minimal disease, 6 per cent. In the Negro group the mortality rate after five years was as follows: far advanced disease, 78 per cent; moderately advanced disease, 40 per cent; and minimal disease, 19 per cent.

In the present series the recovery rate for the Negro is 24 per cent as compared to 69 per cent (table 1) for the white patient. When the two white patient groups are compared there is found recovery in 72.7 per cent of the private patients as compared to 63.6 per cent for the state and city patients. As would be expected, the follow-up on the private patient group is better than in the other group of white patients, that is, 84.2 per cent as compared to 60.8 per cent. The follow-up is exceptionally good on the Negro patients, 82 per cent (table 1). Many of the Negro patients are still ill or are dead and the number of fatalities can be ascertained through the Bureau of Vital Statistics.

The question arises as to whether the difference between the death rate in white and Negro patients is attributable to the difference in the economic status of the two groups. When the two groups of white patients are compared, it can be seen that the economic factor is important for here there is no important racial difference to be considered.

In the early years of the study the indications for pneumothorax were not understood as well as at present. Undoubtedly, many patients were subjected to the closed type of pneumonolysis when they were not satisfactory candidates for the procedure. Thus, late complications were probably higher and the death rate from tuberculosis greater than would be the case under present standards and indications. Several of the patients developed bronchopleural fistulas because of large tension cavities and opaque lungs. At present pneumothorax would not be continued in such patients and they would probably be subjected to pulmonary resection rather than be accepted for the closed form of pneumonolysis.

If pneumonolysis is considered a procedure to be judged by early complications and whether or not the operation is successfully accomplished, the record would indicate that it is a safe operation. The series-mortality rate is 2 patients of the 608 who were subjected to 685 operations. The lung was completely freed of adhesions in 49.5 per cent of the patients and was incompletely freed in 32.1 per cent of the patients. Thoracoscopy followed by inspection only was performed in 18.2 per cent of the patients. The pneumothorax space was lost in 4 of the patients. A record of such low mortality and morbidity would indicate that

all patients with adhesions should be subjected to the closed type of pneumonolysis provided that the patient was originally a suitable candidate for pneumothorax. The belief is strengthened when it is realized that the incidence of late complications are certainly no greater after the closed form of pneumonolysis than without the operation. The incidence of tuberculous and mixed infection empyema was 3.8 per cent when the lung was freed, 9.3 per cent if the lung was freed incompletely, and 7 per cent when adhesions were not disturbed. The incidence of empyema occurring in pneumothorax without pneumonolysis as found by Eglee and Wylie (22) was 7.8 per cent. These writers also summarized the reports of six others who gave the incidence of tuberculous and mixed infection empyema as ranging from 5.8 to 14.4 per cent. Alexander (1), however, reported the incidence as ranging from none in 600 cases to 21 per cent in 151 patients having pneumothorax without pneumonolysis. Even though the mortality rate is higher in the Negro patient, the incidence of complications including empyema is essentially the same as in the white patient.

By definition a complete pneumonolysis is accomplished when the lung is freed completely of all adhesions. A patient with complete pneumonolysis should have the best prognosis for healing of the lung disease. In the private patient group 84.6 per cent (table 1) of these patients are living and well. In the state and city sanatorium group of patients there are 73.3 per cent living and well after complete pneumonolysis. The results of complete pneumonolysis should be compared with results when thoracoscopy only was done. In both white patient groups approximately 60 per cent are living and well. It is not satisfactory to consider the incomplete pneumonolysis group for judgment must be exercised as to whether the resulting collapse will be adequate or inadequate. Many will be no better after incomplete pneumonolysis than after thoracoscopy.

The percentage of patients who were subjected to thoracoscopy without pneumonolysis seems rather high (18.2 per cent). It should be noted, however, that patients were not selected for operation from the standpoint of whether or not the adhesions could be released. If pneumothorax was initiated and the collapse was inadequate because of adhesions, the operation was undertaken even though freeing of the lung seemed impossible by roentgenogram or by fluoroscopy. Thoracoscopy is of definite help to the patient because at times it provides information leading to the early abandonment of an unsatisfactory pneumothorax.

Patients whose lung did not collapse well under proper refill treatments were subjected to operation whether or not adhesions could be demonstrated. Fifteen such cases were encountered and 3 of these patients did not have adhesions. They are included in the group with thoracoscopy without pneumonolysis.

It would seem that the early closed type of pneumonolysis after the initiation of pneumothorax was not carried out consistently (table 3). It was difficult to make known the advantages of early pneumonolysis. There is also the difficulty of promptly obtaining a bed in the general hospital. Occasionally there is a distinct advantage in withholding pneumonolysis for several weeks for adhesions sometimes stretch out. These cases are in the minority. It is true that in some patients the adhesions stretch considerably under careful pneumothorax treat-

ment. It should be pointed out that the force used to stretch an adhesion is made possible by the pneumothorax. However, the intrapleural pressure used is only a small factor in the stretching of most adhesions. The weight of the lung falling by gravity exerted upon the adhesion is the most important factor in the stretching of the adhesion. If the position of the adhesions were known, the patient undoubtedly could be urged to assume the position that will allow the weight of the lung to fall upon the adhesion. This maneuver has been used in the patients who have had incomplete pneumonolyses and occasionally it has proved successful as the lung could be freed completely at subsequent operations. It would appear that this factor could be used in patients who have not yet been subjected to operation. The posteriorly placed adhesions are the ones which most frequently defy release by operation. It is true that most tuberculous disease is posterior in the lung but it is likewise true that most patients, whether they have pneumothorax or not assume the supine position more often than any other position and particularly more frequently than the prone position.

Hemorrhage of varying degrees occurs in every patient subjected to the closed type of pneumonolysis. Some blood must escape from the trocar site into the pleural cavity. Whenever it was necessary to cauterize a blood vessel to stop oozing or brisk bleeding, the case was tabulated under hemorrhage and 28 cases occurred in the series. In no instance did a hemorrhage of more than an estimated 100 cc. occur, nor was a transfusion of blood or plasma ever necessary.

Obliterative pleuritis is thought to develop frequently after free blood has been found in the pleural cavity (23). In the series of 28 patients tabulated as having had hemorrhage at the time of operation, not one developed obliterative pleuritis. In the entire series, obliterative pleuritis occurred in only 3 patients and all the patients operated upon had some blood escape into the pleural cavity.

SUMMARY

1. The open type of pneumonolysis may be indicated when the closed method fails to accomplish a satisfactory collapse of the lung. The open form is a good method of treatment in selected patients.

2. The individual operator is more important than the choice of a one or two piece pneumonolysis instrument or the selection of galvanocautery or electrosurgery as the cutting method.

3. When possible, adhesions should be released and not divided.

4. The indication for closed pneumonolysis is the presence of intrapleural adhesions in a patient who is a candidate for artificial pneumonothorax.

5. Closed pneumonolysis usually is accomplished advantageously in a few days after initiation of the artificial pneumothorax.

6. The closed type of intrapleural pneumonolysis is a safe operation, but its value should not be dissociated from the end results of pneumothorax treatment.

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THE DIAGNOSTIC PROBLEM OF PRIMARY PLEURAL EFFUSIONS[1]

JOSEPH R. KRAFT

INTRODUCTION

The accepted practice at present is to consider all primary pleural effusions as tuberculous or "probably tuberculous" until proved otherwise. It is well recognized that substantial differences in regard to prognosis and treatment exist between tuberculous pleural effusions and the benign effusions of other etiology. Unfortunately, there are available no generally applicable diagnostic methods for the ready identification of the various forms of primary pleurisy. In the absence of diagnostic tests, it was believed that it might be worth while to review a large series of cases of primary pleural effusion with particular attention to the diagnostic problems involved. Such a review constitutes the basis for the present report.

OBSERVATIONS

Composition of Series

One hundred patients with serous pleural effusion observed on the tuberculosis service of Fitzsimons General Hospital, Denver, Colorado, have been reviewed. The patients were admitted to this hospital within a period from May, 1947 through January, 1948. No pleural effusion which appeared in association with, or subsequent to, known pulmonary disease or a surgical procedure was included in this series. Therefore, the cases reviewed may be considered "primary." Only those cases in which observations were made for a minimum of six months in the hospital are included.

Classification

The cases were grouped according to reoentgenographic evidence of parenchymal disease and the results of studies of sputum and pleural fluid after a minimum of six months or more of hospitalization. Group I consisted of 55 cases without evidence of pulmonary lesions or abnormal laboratory findings. Group II consisted of 24 cases without evidence of pulmonary disease but with bacteriologic evidence of tuberculosis. Group III consisted of 21 cases in which evidence of pulmonary disease (with or without positive bacteriologic tests) was detected after the appearance of the effusion.

The series is necessarily a select group as it consists only of American males (97 soldiers and 3 veterans). Sixty-two per cent were in the age group 18 to 21 years and 90 per cent were in the 18 to 30 year old group. The youngest patient was 17 and the eldest, 39 years of age. The series of 100 cases included Negroes and 7 Japanese-Americans (table 1). The great majority of the cases (97 per cent) were transferred from other Army hospitals. Seventy-one per cent of the patients developed their illness while on foreign duty (table 2).

[1] From the Medical Service, Fitzsimons General Hospital, Denver, Colorado.

None of the patients recalled having been exposed to tuberculosis during civil life. Of those with foreign service, only 6 had definite tuberculosis exposure while an additional 9 had had questionable exposure. With the known high prevalence of pulmonary tuberculosis in Germany and the Far East, it may be assumed that these patients were exposed to the disease.

Of the 100 patients in the series, 46 had right-sided effusions and 54 had left-sided effusions. There were only three instances of bilateral effusion. Mantoux

TABLE 1

Racial composition of series

	GROUPS		
	I	II	III
White	45	21	13
Negro	8	1	5
Japanese-American	2	2	3
	—	—	—
Total: 100 cases.	55	24	21

TABLE 2

Incidence of foreign service

	GROUPS		
	I	II	III
Germany	20	8	2
Japan	21	5	3
Korea	6	3	0
Others*	1	1	1
	—	—	—
Total: 71 cases.	48	17	6

* Guam, Laborador, Puerto Rico

and coccidioidin (1:1,000) skin tests were performed on all cases. Seventy-seven reacted positively to the first strength PPD and 23 were positive to second strength. Eight patients reacted positively to coccidioidin (one plus). The pleural fluids of 3 of these patients were positive for tubercle bacilli on guinea pig inoculation. The remainder had never been in any of the areas where coccidioidomycosis is known to be endemic. The coccidioidin skin tests were evaluated as insignificant.

Symptoms

As may be seen in table 3, chest pain, varying from a dull to a sharp, piercing pain, more marked on deep respiration, was the predominant complaint (91 per cent) and was usually associated with fever and malaise (88 per cent). The pain

and the effusion always occurred on the same side of the chest. The associated fever would usually vary from 99 to 103° F. Only one patient had a fever as high as 105° F. He was considered to have an atypical pneumonia with effusion until the presence of tubercle bacilli in the pleural fluid was demonstrated. In the entire series the average duration of fever was eighteen to twenty-one days. A duration of four to six weeks was not uncommon. The fever usually terminated by lysis and did not appear to have been affected by chemotherapy. Seventy-two patients received penicillin, 25 were given sulfonamide alone or in conjunction with penicillin, and 3 were treated with streptomycin.

It was noted that in 15 patients with a fever of 103° F. or higher a gradual drop to a low grade fever followed aspiration of the pleural fluid. Conversely, the low grade fevers seemed relatively unaffected by aspiration of the fluid.

Eighty-two of the 100 patients lost weight during their illness. The amount of loss varied from 5 to 20 pounds. The loss of weight occurred principally during the acute or febrile phase of the illness. This weight was usually regained quite readily following termination of the acute phase.

TABLE 3

Subjective symptoms

TOTAL		GROUPS		
		I	II	III
94	Chest pain	53	22	19
88	Malaise and fever	48	20	20
25	Cough and sputum	14	6	5
12	Dyspnea	6	4	2
82	Weight loss	40	22	20

It was particularly interesting to note the relatively insidious onset of symptoms in primary pleural effusions. A careful history of subjective symptoms, with particular emphasis on onset of chest pain, revealed that 73 per cent experienced the onset of symptoms one or more weeks prior to hospitalization. The initial onset was usually one of intermittent, transitory chest pain, occasionally "sharp", but usually dull. Increased chest pain plus fever and malaise would usually force the patient to seek medical aid. The duration of symptoms before seeking medical care was two to seven days in 27 per cent of the group; eight to fourteen days in 30 per cent; fifteen to twenty-one days in 20 per cent; and twenty-two to twenty-eight days in 15 per cent (table 4). Four patients had had symptoms for three months prior to hospitalization and one patient had been ill for five months. It was also noted that in 10 patients the pleural effusions were discovered by routine chest roentgenograms. In three of these no history of any symptoms could be elicited.

Of the 100 cases reviewed, in only 10 were the initial subjective symptoms of chest pain sufficiently severe to warrant seeking prompt medical aid. Roentgenographic examinations at the time of onset of the pain were negative. Sub-

sequent roentgenographic examinations, following recurrence or aggravation of symptoms, revealed the presence of an effusion. Four of these cases were in group I and three were in groups II and III, respectively.

In only 11 instances was it possible to note the relative progression of effusion. Three of these patients developed effusion during hospitalization for other causes. Symptoms had been present for two to four days before the first roentgenographic examinations. These revealed haziness of the lateral lower lung field at the level of the sixth or seventh anterior rib with blunting of the costophrenic sulcus or with definite evidence of fluid. Serial roentgenographic and fluoroscopic examinations revealed that the maximum amount of fluid was demonstrated in these cases between the fourteenth and the twenty-first days after the onset of symptoms. Of these 11 cases, 5 were in group I, 4 in group II, and 2 in group III.

TABLE 4

Duration of symptoms prior to hospitalization

PER CENT	TIME	GROUPS		
		I	II	III
27	2 to 7 days	17	6	4
30	8 to 14 days	14	10	6
20	15 to 21 days	11	5	4
15	22 to 28 days	8	2	5
5	one month, plus	3	0	2
3	no symptoms	2	1	0
100	Total.....	55	24	21

With the triad of chest pain, fever and malaise, and the roentgenographic findings of haziness in the lateral lower lung field, a preliminary diagnosis of atypical pneumonia with effusion was made in 7 of the 11 cases.

Laboratory Data

In the 90 cases which were aspirated, the fluid was generally described as clear amber (91 per cent) or cloudy amber (9 per cent). There were no instances of purulent or bloody fluid in this series. The specific gravity of the fluid was typical of exudation (1), *i.e.*, usually above 1.015 and invariably above 1.012.

Of the effusions aspirated, 51 were tested by guinea pig inoculation with 18 positive results (35 per cent); 30 effusions were tested by culture alone of which 2 were positive for tubercle bacilli. It should be noted that of those tested by guinea pig inoculations 24 were, in addition, tested by culture. Nine fluids were positive for tubercle bacilli with both tests. Cultures of 80 fluids for microorganisms other than *M. tuberculosis* were uniformly inactive. It should be noted that the animal inoculations and cultures reported above were single tests. Repetition of these examinations would have been highly desirable.

Regardless of method, the erythrocyte sedimentation rate was markedly elevated at the onset of the effusion. The elevation would persist long after the

acute febrile phase of the illness had subsided. In many cases the elevation persisted the entire period of observation (six to eight months) prior to transfer of the patient to a Veterans Hospital. It was noted that the course of the sedimentation rate seemed directly related to the regression of the pleural fluid or fibrinous pleurisy. All patients with a minimal fibrinous pleurisy which was stable roentgenographically for two or more months within six months of onset had sedimentation rates within normal limits. This was also true of 5 patients who subsequently developed parenchymal lesions (table 6, cases 7, 9, 16, 17, 18).

Sixty-two per cent of the patients had total leucocyte counts which were within normal limits (2). In every instance the differential counts were within the normal range, including the determinations in the 28 patients with a mild

TABLE 5

Laboratory examinations of pleural fluid and sputum

	GROUPS		
	I	II	III
*Specific gravity above 1.015	41	20	14
*Specific gravity below 1.015	2	1	2
Lymphocytes predominate	42	19	14
Polymorphs predominate	1	2	2
Positive guinea pig inoculation for *M. tuberculosis*	0	15	3
Positive culture *M. tuberculosis*	0	2	0
*No aspiration	7	1	2
*Results unknown	5	2	3
Sputum positive for *M. tuberculosis*	0	8	8
	55	24	21

* Total—100

leucocytosis (up to 12,500 per cm^2). The erythrocyte and hemoglobin determinations were all within normal limits.

Occurrence of Parenchymal Disease

In this series of 100 cases, 21 subsequently developed roentgenographically demonstrable parenchymal disease following the onset of the pleural effusion (table 6). It should be emphasized that no instance of effusion concomitant with known parenchymal disease or primary complex was included in the series. In all but 3 cases roentgenograms obtained two to twenty-four months before the onset of effusion were available. In the 3 without previous films, it was possible to observe the parenchymal lesions roentgenographically following the onset of effusion.

All of the parenchymal lesions were minimal in extent except two (cases 6 and 7) which were moderately advanced. The apical lesions were typical of reinfection pulmonary tuberculosis. Ten lesions were suggestive of a primary parenchymal focus. In only 2 of these cases (7 and 19) was an associated hilar

adenopathy typical of a primary complex noted. In all of the patients except cases 7, 13, and 17, detailed below, the parenchymal lesions were demonstrated roentgenographically during a regimen of bed-rest.

TABLE 6

The occurrence of parenchymal disease (Group III)

CASE NUMBER	LAST NORMAL ROENTGENOGRAM	CHRONIC FIBRINOUS PLEURISY AT SIX MONTHS (OR AS INDICATED)	ROENTGENOGRAPHIC ONSET OF LESION IN MONTH AFTER ONSET OF FLUID	FLUID FOR *M. tuberculosis*	SPUTUM EXAMINATION FOR *M. tuberculosis*; MONTH FOUND	SITE OF LESION
	months					
*(1)	2	m[2]	2	negative	+ (8)	Apex (right)
(2)	6	M	10	+	negative	RLL
(3)	3	M	9	negative	+ (6)	RLL
(4)	12	M	3	—	negative	Apex (right)
(5)	18	m[5]	4	—	negative	Apex (right)
(6)	—	M	6	negative	+ (5)	Apex (right)
(7)	12	m[2]	6	negative	negative	RUL
(8)	18	M	8	negative	negative	LLL
(9)	6	m	6	—	negative	LLL
(10)	23	M	6	—	+ (8)	LLL
(11)	15	M	5	negative	negative	LUL
(12)	18	m	6	negative	negative	Apex (right)
(13)	—	M	10	negative	negative	RUL
*(14)JA	8	m	6	negative	+ (5)	RLL
(15)JA	10	M	3,6	negative	+ (5)	Apex (bilateral)
(16)JA	2	m	6,8	negative	negative	Apex (bilateral)
(17)N	22	m	8	negative	+ (9)	Apex (left)
(18)N	9	m	6	negative	+ (8)	Apex (right)
(19)N	12	M	6	negative	+ (6)	RLL
(20)N	24	M	7,9	+	+ (9)	Apex (bilateral)
*(21)N	—	m[4]	3	+	negative	Apex (bilateral)

M: moderate chronic pleurisy extending laterally up to the fourth anterior rib
m: minimal chronic pleurisy extending laterally up to the sixth anterior rib or less
JA: Japanese-American
—: results unavailable
N: Negro
*(): bilateral effusion

Case 7: The patient had been hospitalized three months for atypical pneumonia with effusion. The fluid negative for tubercle bacilli and a chronic minimal pleurisy persisted. Three months later, at the separation center, a minimal parenchymal focus with associated hilar adenopathy was visible on a roentgenogram of the chest. Regression of the lesion was noted after four months of bed-rest.

Case 15: After six months hospitalization for primary pleural effusion, the patient went A.W.O.L. for six weeks. Upon return, a minimal parenchymal focus was demonstrable on the chest roentgenogram.

Case 17: The patient was hospitalized for primary pleural effusion. The fluid, which was negative for tubercle bacilli, disappeared almost completely after six months' hospitalization. No parenchymal lesion was noted at this time but after the return of the patient from

a convalescent furlough (forty-five days) a minimal apical lesion was discovered. Tubercle bacilli were present in the sputum. The lesion progressed to cavitation.

Three patients had bilateral effusions with initial onset on the right and subsequent contralateral effusion at two and one-half, three, and four months, respectively. One of the patients (case 21) developed bilateral, minimal apical lesions three months after the onset of the pleurisy, a peritoneal effusion six months later, and tuberculous meningitis confirmed by culture eight months after the onset of the pleural effusion. This was the only case comparable to those reported by Fernandes (2) who believes that bilateral pleural effusion is frequently a manifestation of miliary tuberculosis.

The results of the bacteriologic examinations of the sputum are presented in table 6. It is granted that the finding of a parenchymal lesion did prompt repeated and intensified studies of the sputum. Bronchoscopy of all patients whose sputum contained *M. tuberculosis* revealed no endobronchial disease. All of the parenchymal lesions were initially noted upon the same side as the effusion, including 3 of the instances of bilateral effusions. In the remaining instance (case 21) with bilateral lesions, it was impossible to determine which lesion developed first, for both were noted upon the same roentgenographic examination.

Pathogenesis

Israel and Long (3) have reported that primary tuberculous infection in adults, as evidenced by a change in tuberculin sensitivity from negative to positive, may be followed after a short interval by an attack of pleurisy.

Erwin (4) in 1944 postulated reasons why an intrathoracic primary complex might be expected to produce direct infection of a highly allergic pleura with tubercle bacilli penetrating either from a subpleural parenchymal focus, or from a caseous tracheobronchial lymph node.

Thompson (5) accepts this theory and refutes the theory of post-primary dissemination by the blood stream on the basis of his observations on 190 cases.

The Erwin theory, especially as elaborated by Thompson, may quite well be the explanation of pathogenesis of many of the cases in the present series. As previously noted, all of the patients had positive tuberculin skin tests. A good deal of information favoring or refuting Erwin's theory could have been obtained from the present series had the history of tuberculin conversion been known.

Prognosis

It is granted that the time period of observation of the cases reviewed is short and that much must be speculated in regard to the eventual course of the cases. It should be noted that a long term survey of primary pleural effusion cases (including cases reviewed) is currently being evaluated by Dr. Wm. H. Roper, Director of Minimal Tuberculosis Research, Fitzsimons General Hospital (6). However, it is significant to note that 21 per cent of the present series developed pulmonary tuberculosis within one year (average 6.2 months) after the onset of pleural effusion. In regard to prognosis, this percentage may be compared with

Thompson's (7) findings where 12 per cent of 190 patients with pleural effusion developed a parenchymal lesion within the first year and a total of 25 per cent within five years.

Bonilla's (8) report may also be applied with some concern to the cases reviewed, for in his group of 40 patients with pleural effusions without any pulmonary infiltration, the parenchymal lesions developed in the whites in an average of 38 months, and in the Negros 21.3 months after the pleurisy.

Jones and Dooley (9) demonstrated that pleural effusion may be a manifestation of extrapulmonary tuberculosis, especially in the Negro. They also emphasized the necessity for a guarded prognosis in Negro patients with effusion, for in their series of 99 cases followed for more than one year 15 per cent eventually died of tuberculosis which was principally of an extrapulmonary form. In the total cases reviewed in the present report, only one presented evidence of extrapulmonary tuberculosis during the short time of observation. It was an instance of renal tuberculosis in a Negro of group I.

COMMENT

Although it is generally accepted that a primary pleural effusion is frequently a manifestation of tuberculosis, the insidious onset, chest pain, fever and malaise do at times present problems of differential diagnosis.

In this series of 100 cases, 24 were originally considered to have nontuberculous diseases, including pneumonia with effusion, bronchitis, and malaria. The diagnosis of atypical pneumonia with effusion was most common. This error occurred especially when the patient presented himself after a relatively short duration of symptoms and only a minimal effusion was demonstrable roentgenographically at the onset of the illness. Particular reference is made to the cases of atypical pneumonia with effusion as cited by Turner (10). Several of his examples were comparable with almost any one of the 27 patients of the present series who had had symptoms for less than one week. Of the 24 cases that were originally considered to have diseases other than tuberculosis of the pleura, 13 were in group I, 7 in group II, and 4 in group III.

Examination of the fluid may give much information in regard to etiology and at times may establish the diagnosis (11 to 17). The importance of a positive finding of tubercle bacilli on culture or guinea pig inoculation of the pleural fluid is evident. Nevertheless, the relatively low incidence of successful isolations of tubercle bacilli from the pleural fluids of the present series (35 per cent) and in other reports (5, 18, 19) serves to emphasize that the diagnosis for the most part depends upon careful differential diagnosis and clinical experience.

Since many of the effusions were tested at other hospitals, the techniques employed are unknown. At the laboratories of Fitzsimons General Hospital, according to Fisher (20), the centrifuging of large amounts of pleural fluid followed by guinea pig inoculation of the resulting sediment produces the most favorable results. Granting that with a uniformly optimal technique, the results might have been somewhat better, the problem is not appreciably lessened.

The necessity, because of the lack of absolute certainty, to designate those in-

fections in which the etiology has not been demonstrable, *e.g.* group I, as "probably tuberculous" occurs. The possibility that these cases may represent other diseases, *e.g.*, atypical pneumonia or rickettsial infections, cannot be completely excluded.

As noted in the cases reviewed, it is impossible to foretell which infections may progress to the development of parenchymal or other lesions. Hence, in the light of current knowledge, treatment must be initiated empirically.

The treatment of primary pleural effusion had dual objectives. The maximum removal of fluid in an effort to limit the chronic fibrinous and eventually fibrous pleurisy to a minimum may be cited as one objective. The other is bed-rest for the underlying tuberculous lesion, whether it be visible or invisible on roentgenographic examination. This bed-rest may be considered as prophylaxis rather than therapy. It is recalled that, in the majority of the 21 cases with lesions, the latter appeared while the patients were on bed-rest. Three cases were also cited that demonstrated the dangers of too early activity. Bonilla (8) noted in his series that the patients whose lesions became known during rest had a much more favorable prognosis than those whose lesions were detected during periods of physical activity.

It is believed that at least one year of good sanatorium care with complete bed-rest is the minimum amount of treatment which is advisable for a patient with primary pleural effusion. Periodic roentgenographic examination during the subsequent four years is of equal importance as recommended by Thompson (7). As noted above, a normal erythrocyte sedimentation rate within the first six months of rest does not necessarily preclude the subsequent development of a parenchymal lesion. Moreover, a normal value for the sedimentation rate is not a trustworthy guide by which to determine the proper duration of treatment.

The responsibility upon the physician, especially in civilian life, to regard an effusion as "probably tuberculous" and to initiate recognized treatment is tremendous. It entails study of possibilities versus probabilities in which often, if not invariably, the patient will make the final decision as indicated by the response and cooperation with treatment.

The degree of cooperation is usually directly proportional to the degree of certainty of diagnosis of the physician. The patient, when there is question or choice of diagnosis, will invariably apply the diagnosis of lesser consequence to his case.

It is relatively easy to practice didactic medicine in the Armed Forces. Among civilians, however, a period of long continued bed-rest creates a terrific economic problem. The financial catastrophe related to prolonged hospitalization frequently immediately overshadows the subsequent possibilities and probabilities of a course of disease to the patient. The physician must, with supreme effort, divorce himself from this influential factor in making the diagnosis and prescribing the necessary treatment.

There is still much to be learned about primary pleural effusions. A means of definitely establishing etiology and thereby removing the doubt from both the physician and the patient is urgently needed.

SUMMARY

1. A review of 100 cases of primary pleural effusion is presented. Twenty-one of the group developed pulmonary lesions and one developed renal tuberculosis within 6.2 months of onset.

2. The insidious onset of subjective symptoms was striking. Seventy-seven per cent of the series had symptoms for seven or more days prior to seeking medical aid and in ten instances the effusion was first detected on a routine roentgenographic examination.

3. It is apparently impossible to predict from the findings during the stage of the effusion whether pulmonary or other lesions will subsequently occur.

4. In the absence of satisfactory diagnostic tests, certain cases of pleural effusion must be classified as "probably tuberculous," although they may represent instances of primary atypical pneumonia. In general, such cases should be treated in the same way as the cases in which the tuberculous etiology is established.

5. The treatment recommended for primary pleural effusions consists of a period of sanatorium care (usually one year) followed by clinical, bacteriologic and roentgenographic examinations.

SUMARIO

La Importancia del Diagnóstico en el Derrame Pleural Primario

1. Esta reseña versa sobre 100 casos de derrame pleural primario. Veintiuno del grupo manifestaron lesiones pulmonares y uno manifestó tuberculosis renal en término de 6.2 meses de la iniciación.

2. Fué notable la insidiosa iniciación de los síntomas subjetivos. Setenta y siete por ciento de los enfermos habían tenido síntomas desde siete o más días antes de consultar al médico y en 10 casos se descubrió el derrame por primera vez al verificar un examen radiográfico de los corrientes.

3. Parece aparentemente imposible predecir por los hallazgos del período de derrame si se presentarán o no luego lesiones pulmonares o de cualquier otro género.

4. A falta de satisfactorias pruebas de diagnóstico, hay que clasificar ciertos casos de derrame pleural como "probablemente tuberculosos," aunque pueden representar neumonía atípica primaria. En general, esos casos deben ser tratados lo mismo que los de etiología tuberculosa establecida.

5. El tratamiento recomendado para los derrames pleurales primarios consiste en un período de asistencia sanatorial (por lo general un año), seguido de exámenes clínicos, bacteriológicos y radiográficos.

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PULMONARY EMPHYSEMA AND VENTILATION MEASUREMENTS IN ONE HUNDRED ANTHRACITE COAL MINERS WITH RESPIRATORY COMPLAINTS[1, 2]

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INTRODUCTION

Anthracosilicosis (miner's asthma) is an occupational disease, characterized by silicotic pulmonary fibrosis, excessive retention of carbonaceous material and emphysema (1). Emphysema has been described as one of the most frequent gross findings, and the principal histologic change is a fibrous hyperplasia along lymph channels which occurs early and increases in certain areas to form nodules. Outstanding clinical features generally observed in anthracosilicosis include progressive dyspnea, accentuated by intercurrent infections, symptoms of failure of the pulmonary circulation and disability. In some cases the chest roentgenogram may be essentially normal, yet clinically there is a marked degree of disability. Emphasis has been placed on the diagnosis of silicosis from a correlation of all the facts (2), symptoms, signs, occupational history and laboratory and roentgen observations. In the present study information has been obtained on the ventilation impairment in anthracosilicosis from quantitative measurements of residual air, vital capacity, resting minute ventilation and maximal breathing capacity. These observations have been made on 100 unselected patients with respiratory complaints and a history of long exposure inside the mines. The pulmonary function and roentgenographic findings have been compared.

METHODS

The residual air was determined by the open circuit oxygen breathing method previously described (3). In the present study, however, a demand type regulator supplied the oxygen, replacing the constant flow regulator and rubber bag used in the original assembly. The subject was allowed to breathe 99.6 per cent oxygen for a seven minute period, during which all expired air was collected in a calibrated Tissot type spirometer. At the end of the seven minute period of oxygen breathing a sample was taken for the alveolar nitrogen determination. The dead space in the valve arrangement used for switching from air breathing to oxygen breathing and for alveolar nitrogen sampling was 65 cc. Duplicate residual air determinations were routine and required to check within 100 cc. in all cases except a few with a far advanced degree of emphysema where a tolerance of not over 5 per cent was permitted.

Maximal breathing capacity (MBC), vital capacity and supplemental or reserve air were determined with a modified Benedict-Roth metabolism apparatus (4). The predicted total lung volume was calculated from the height, age and sex (5). The bell of the apparatus has a diameter of 22.9 cm. and a volume of 13.5 L. The increased volume reduces the amplitude of the vertical movement of the bell with each breath and decreases the resistance for rapid

[1] From the Cardio-Respiratory Laboratory, Barton Memorial Division of Jefferson Medical College Hospital and the Department of Medicine of Jefferson Medical College.

[2] Investigation aided by a research grant of the Anthracite Health and Welfare Fund of Jefferson Medical College.

and deep breathing. The breathing resistance was further reduced by employing large tubing with an inside diameter of 2.9 cm. in conjunction with the high velocity directional valve used to prevent excessive accumulation of CO_2. Multiple measurements were obtained in every case, each being a twelve second run, with varying rates and depths of breathing. The values reported were the highest obtained. Measurements made on trained subjects with the above apparatus, which was designed to reduce resistance to a minimum for the MBC determinations, check closely with values obtained when employing the 100 L. Douglas bag and the high velocity directional valve. Two writing pens are used on the electrically driven (two speed) kymograph, one to record amplitude and rate of breathing and the other to add the total volume. The regression formula used for calculating the predicted values was: MBC L/min. = 86.5 − (0.522 × age in years) × B.S.A. (sq. m.) (6). All ventilatory volumes are measured as saturated gas at 37°C. and the prevailing barometric pressure.

Roentgenograms of the chest were taken at full inspiration and at full expiration in postero-anterior view. Routine fluoroscopy was done on all the patients.

All residual air measurements were made with the patients in a fasting and resting state lying supine and as flat as was comfortable for the individual. No medication was given to the patients. All MBC determinations were made in the standing position.

MATERIAL FOR STUDY

All of the patients were male anthracite coal miners with a long history of exposure to dust in mines. These miners were sent to the Barton Memorial Division of Jefferson Hospital for a study sponsored by the Anthracite Health and Welfare Research Fund of Jefferson Medical College. Patients were admitted to the Barton Memorial Hospital in order of the date of application for study, hence there was no selection by the laboratory staff except that subjects with proved pulmonary tuberculosis were omitted. However in 5 cases active tuberculosis was diagnosed after study (table 1). All but one of the patients complained of dyspnea. Chest pain, cough and expectoration, weakness, loss of weight and hemoptysis were the other most common complaints. In some patients dyspnea was present only during the course of minor respiratory infections, while in others there was a history of cyclic recurrence of the symptoms. All degrees of dyspnea were observed from the most profound, even during bedrest, to slight exertional dyspnea after exercise. There was no evidence of cardiac decompensation in this group at the time of study. However, two patients were known to have cardiac disease with electrocardiographic evidence of myocardial damage (table 1).

RESULTS

A quantitative indication of the degree of emphysema was obtained by expressing the residual air as per cent of the total lung volume (residual air + vital capacity). This is a physiological comparison of the relationship of the volume of residual air on the mixing and dilution factor with respect to the vital capacity and total lung volume. It is a more accurate measurement of pulmonary function impairment than the changes in residual air from the predicted, in which the wide variations in vital capacity are not correlated. Furthermore the total lung volume varied less from the predicted than either residual air or vital capacity measurements for the entire group (table 1). Accordingly the degree of emphy-

TABLE 1

Lung volume ventilation measurements, inside mine exposure time, and roentgenographic stage of silicosis in 100 cases of anthracosilicosis arranged in order of increasing residual air per cent of total lung volume

PATIENT	AGE	HEIGHT	BODY SURFACE AREA	EXPOSURE INSIDE MINES	X-RAY STAGE	RESIDUAL AIR		VITAL CAPACITY		MAXIMAL BREATHING CAPACITY		VENTILATION	BREATHING RESERVE OF MAXIMAL BREATHING CAPACITY	TOTAL LUNG VOLUME CHANGE FROM PREDICTED
						Observed	Total Lung Volume	Observed	Decrease From Predicted	Observed	Decrease From Predicted			
I. Normal or no pulmonary emphysema														
	Yrs.	cm.	sq.m.	Years		cc.	per cent	cc.	per cent	L./minute	per cent	L./minute	per cent	per cent
J. C.	45	166	1.64	19	2	500	11.0	4,050	0.0	101	2.0	8.9	91.4	−13.8
J. P.	52	176	2.05	26	2	720	13.8	4,500	0.0	66	46.0	7.8	88.2	− 8.8
L. C.	40	173	1.84	18	3	879	16.6	4,400	0.0	94	21.4	6.4	93.2	− 9.0
W. R.	53	167	1.80	38	3	813	16.9	4,000	0.0	65	39.0	8.4	87.2	− 5.6
R. P.	53	170	1.70	25	3	543	19.4	2,280	43.0	43	57.3	6.5	85.0	−46.7
F. R.	54	171	2.08	40	2	898	19.6	3,680	9.7	107	12.4	10.0	90.6	−14.9
P. H.	44	172	1.72	28	3	975	20.2	3,682	6.0	81	29.0	6.9	91.2	−11.1
J. M.	44	175	1.89	25	2	1,091	20.4	4,270	0.0	103	5.0	6.7	92.3	− 5.0
H. Q.	47	170	1.74	30	2	1,181	20.8	4,500	0.0	112	0.0	7.9	93.0	+ 6.5
R. Z.	46	173	1.75	31	3	1,111	20.8	4,200	0.0	67	38.8	10.2	85.0	− 3.8
J. F.	57	174	1.95	36	2	1,254	21.8	4,500	0.0	96	13.7	6.8	92.8	+ 3.3
J. F.	41	170	1.80	20	3	1,219	22.3	4,250	0.0	90	30.0	9.7	89.2	+ 3.2
A. B.	61	170	1.75	26	3	887	22.5	3,045	24.0	50	48.0	7.0	86.0	−26.0
G. P.	44	165	1.78	24	1	1,033	23.1	3,440	8.3	87	23.5	8.1	90.6	− 8.4
Av.....	49	171	1.82	28		936	19.2	3,910	6.5	83	26.1	8.0	89.6	−10.0
II. Slight degree of emphysema														
B. G.	52	164	1.63	30	2	1,280	25.5	3,730	0.0	97	0.0	9.7	90.0	+ 2.5
J. K.	48	172	1.88	20	2	1,405	26.0	4,000	8.0	38	67.0	8.8	75.8	− 5.7
H. S.	55	173	2.13	35	3	1,014	26.1	2,870	31.2	75	39.4	7.8	89.4	−29.5
P. V.	61	163	1.77	37	3	1,460	26.3	4,100	0.0	59	41.0	9.3	81.6	+15.3
J. S.	71	175	1.76	35	1	1,521	26.4	4,250	0.7	74	14.6	8.1	89.2	+ 2.0
G. K.*	65	158	1.54	37	2	1,123	26.8	3,115	8.5	47	42.0	13.3	71.7	− 4.8
M. E.	51	172	1.76	30	3	1,215	27.2	3,260	20.8	44	58.2	9.0	79.6	−19.7
A. Y.	51	168	1.69	26	1	1,454	28.1	3,720	0.0	68	31.0	8.4	87.7	+12.9
P. C.	43	157	1.60	23	3	1,023	29.2	2,490	25.2	48	53.0	7.5	83.2	−20.1
A. T.	67	169	1.81	47	3	1,469	30.1	3,400	14.1	67	28.7	10.1	85.2	− 7.1
C. S.	40	168	1.83	24	2	1,562	31.8	3,350	14.1	93	22.5	8.6	90.8	− 4.8
J. R.*	65	172	1.56	35	3	1,317	31.8	2,820	31.8	35	57.4	8.5	75.8	−24.0
J. G.	45	178	1.92	31	3	1,246	31.9	2,600	40.2	84	31.0	7.8	90.6	−33.6
F. S.	58	173	1.97	34	3	1,620	31.9	3,430	17.4	53	52.3	9.2	79.4	− 8.2
S. V.	51	171	1.96	37	1	1,547	32.0	3,300	18.9	88	17.7	11.8	86.6	− 9.6
J. A.	60	164	1.81	36	3	1,680	32.4	3,500	3.6	47	53.6	8.0	83.0	+ 7.9
A. S.	56	168	1.64	25	1	1,226	32.4	2,500	34.7	97	0.0	7.7	91.8	−28.9
A. S.	49	178	1.67	27	3	1,380	33.0	2,780	37.4	51	50.0	9.4	81.4	−29.0
L. T.	42	189	2.05	21	3	2,140	33.1	4,320	13.4	89	29.1	9.5	87.8	− 2.7

TABLE 1—*Continued*

Patient	Age	Height	Body Surface Area	Exposure Inside Mines	X-Ray Stage	Residual Air		Vital Capacity		Maximal Breathing Capacity		Ventilation	Breathing Reserve of Maximal Breathing Capacity	Total Lung Volume Change from Predicted
						Observed	Total Lung Volume	Observed	Decrease From Predicted	Observed	Decrease From Predicted			
II. Slight degree of emphysema—*Continued*														
	Yrs.	*cm.*	*sq.m.*	*Years*		*cc.*	*per cent*	*cc.*	*per cent*	*L./minute*	*per cent*	*L./minute*	*per cent*	*per cent*
J.V.	55	166	1.94	25	3	1,615	33.4	3,225	13.2	85	24.6	9.4	89.5	− 1.8
V.V.	54	168	1.73	33	3	1,700	34.4	3,230	20.8	43	57.4	7.2	83.8	− 9.5
P.Y.	45	181	1.84	30	3	1,578	34.8	2,960	35.0	70	40.0	10.3	85.4	−25.0
R.W.	60	173	1.74	40	3	1,073	34.9	2,080	50.2	81	16.0	9.0	89.0	−42.8
Av.....	54	170	1.78	31		1,420	30.8	3,270	19.1	66	36.3	9.0	85.0	−11.5
III. Moderate degree of emphysema														
S.M	58	178	1.87	32	3	1,267	35.3	2,340	47.5	81	22.8	10.4	86.7	−38.6
J.L.	64	175	1.80	35	3	1,405	35.8	3,520	44.5	50	47.9	7.7	84.7	−34.0
P.C.	56	167	1.71	35	2	1,710	36.2	3,010	21.6	81	17.2	8.5	88.8	− 9.7
F.M.	53	170	1.73	38	2	1,945	36.3	3,420	15.6	66	35.7	9.5	85.6	+ 1.0
T.M.	52	172	1.71	27	3	1,275	36.7	2,210	46.3	29	72.4	8.0	72.6	−36.0
J.B.†	53	173	1.93	43	2	2,080	36.8	3,600	12.6	84	26.3	10.7	73.3	+ 4.2
J.S.	45	162	1.58	24	3	1,100	37.3	1,850	48.4	24	76.0	7.9	67.0	−37.8
V.B.	43	180	1.74	25	3	1,282	37.5	2,140	53.0	43	61.8	7.9	81.4	−43.0
J.K.	55	161	1.69	28	3	1,278	37.6	2,120	40.0	27	73.5	6.9	87.3	−40.7
A.G.	59	179	1.88	42	3	2,267	37.8	3,740	16.7	56	46.5	7.3	87.2	+ 1.1
A.N.	72	159	1.69	28	3	1,824	38.6	2,910	19.7	54	35.2	9.7	82.0	− 1.0
D.R.	54	175	1.80	35	3	1,634	38.6	2,600	39.4	32	69.6	10.8	63.2	−25.2
A.S.	45	176	1.78	22	3	1,807	38.6	2,870	33.2	45	59.5	10.3	77.8	−18.6
M.K.	60	166	1.48	27	3	1,066	38.8	1,675	57.0	28	65.9	8.5	69.6	−47.2
T.P.	58	177	1.89	33	3	1,673	38.8	2,640	39.2	41	61.6	10.7	74.0	−25.6
A.H.	59	166	1.74	42	2	1,700	39.0	2,670	29.7	39	60.5	7.8	80.0	−13.3
C.B.	50	165	1.76	27	3	1,736	39.2	2,700	28.0	43	60.0	7.4	83.6	−11.8
C.H.	48	178	2.08	28	1	2,503	39.6	3,800	14.4	61	44.5	9.8	83.6	+ 7.6
P.A.	59	172	1.62	44	3	2,050	39.8	3,115	25.0	42	53.0	7.5	84.5	− 5.4
J.H.	51	166	1.68	36	3	1,810	39.8	2,740	46.0	85	18.9	8.6	89.4	−14.1
J.P.	61	167	1.64	45	2	1,590	41.0	2,290	40.6	48	58.0	7.8	83.4	−23.0
G.T.*	47	169	1.61	16	3	1,737	41.1	2,480	37.4	43	56.7	8.7	81.4	−19.6
K.K.	43	174	1.75	15	3	1,357	42.6	1,820	57.0	32	71.2	5.7	83.4	−42.3
J.U.	62	165	1.75	43	2	2,141	42.7	2,878	23.0	32	66.3	10.8	66.3	+ 1.0
J.C.	46	165	1.84	26	3	1,835	42.9	2,450	26.5	47	58.4	8.1	83.0	−17.6
J.P.	65	157	1.42	44	3	1,654	43.3	2,208	33.5	30	60.5	6.1	79.6	−13.7
B.D.	49	169	1.71	30	3	1,676	43.6	2,160	45.5	40	61.7	7.4	81.5	−27.5
A.H.	60	174	1.83	25	1	2,071	44.0	2,661	37.2	35	65.3	7.1	80.0	−15.5
E.M.	46	164	1.51	20	3	1,067	44.4	1,350	63.5	22	76.5	6.7	69.6	−50.6
J.S.	54	182	1.91	35	1	2,546	44.8	3,140	32.6	34	69.7	7.4	78.2	− 7.5
Av.....	54	170	1.73	32		1,703	39.6	2,637	35.8	46	55.1	8.4	79.6	−20.1

TABLE 1—*Concluded*

Patient	Age	Height	Body surface area	Exposure inside mines	X-ray stage	Residual air: Observed	Residual air: Total lung volume	Vital capacity: Observed	Vital capacity: Decrease from predicted	Maximal breathing capacity: Observed	Maximal breathing capacity: Decrease from predicted	Ventilation	Breathing reserve of maximal breathing capacity	Total lung volume change from predicted
	Yrs.	*cm.*	*sq.m.*	*Years*		*cc.*	*per cent*	*cc.*	*per cent*	*L./minute*	*per cent*	*L./minute*	*per cent*	*per cent*
IV. Advanced degree of emphysema														
J. V.	57	168	1.70	36	2	2,212	45.2	2,680	31.0	42	56.7	7.4	81.6	− 5.2
C. T.	53	174	1.92	34	2	2,410	46.5	2,780	34.5	30	73.5	8.9	70.3	− 7.3
L. M.*	42	169	1.51	24	3	1,626	48.2	1,750	55.2	35	65.0	10.9	68.6	−31.6
H. F.	62	175	1.67	25	3	1,970	48.3	2,120	50.7	25	72.2	7.7	69.2	−28.0
J. Y.*	63	158	1.54	37	3	2,325	48.4	2,480	30.3	29	64.4	8.1	72.0	+ 2.0
S. F.	64	171	1.61	42	3	1,869	48.7	1,970	52.3	23	73.2	7.9	65.7	−29.9
J. B.	42	172	1.85	27	3	2,047	48.8	2,150	47.8	42	64.8	8.5	77.7	−23.0
J. M.	48	167	1.85	23	3	2,373	49.0	2,450	37.0	26	77.0	7.7	70.4	− 5.4
J. K.	62	173	1.63	41	3	2,494	51.0	2,400	42.9	30	65.8	10.6	64.6	−11.6
J. N.	46	169	1.64	25	3	1,418	51.0	1,360	65.0	26	74.2	8.4	69.2	−46.1
G. S.	40	175	1.90	23	3	3,440	51.0	3,290	23.2	56	54.4	9.0	84.0	+18.9
J. B.	53	175	1.86	29	2	2,898	51.8	2,700	37.0	32	67.0	7.3	77.2	− 2.0
A. J.	47	171	1.73	26	3	2,421	52.0	2,230	46.2	27	80.0	7.5	74.0	−13.9
V. P.	44	176	1.68	24	3	2,370	52.0	2,280	27.4	27	74.8	6.6	75.6	−20.7
J. S.	51	164	1.66	20	3	1,517	52.5	1,400	63.2	18	81.6	6.4	66.8	−41.3
W. R.	53	167	1.82	32	3	3,015	53.7	2,610	35.6	31	71.0	8.9	71.4	+ 5.0
G. C.	46	182	1.90	23	3	2,199	54.3	2,620	43.6	34	71.0	5.5	84.0	−21.8
F. M.	57	178	1.69	30	3	2,933	55.8	2,420	54.5	74	22.5	10.2	86.2	− 8.8
Av.	52	171	1.73	29		2,305	50.4	2,315	43.3	34	67.0	8.2	73.8	−15.2
V. Far advanced degree of emphysema														
J. S.	45	168	1.53	24	3	1,753	55.8	1,410	64.0	22	77.5	7.0	68.0	−38.7
R. O.	57	174	1.71	37	3	2,068	56.6	1,598	62.3	24	75.4	6.1	71.5	−31.5
L. Q.	51	177	1.72	25	3	2,250	57.0	1,689	61.6	21	79.0	6.3	70.0	−32.3
H. M.	60	173	1.88	38	2	3,117	57.5	2,335	44.3	29	71.8	8.3	71.4	− 1.4
R. S.	57	164	1.54	31	2	3,330	57.5	2,410	40.0	29	66.0	8.6	70.4	+17.0
G. P.	51	172	1.77	36	1	2,777	57.8	2,020	51.0	27	74.6	7.6	70.4	−12.0
J. B.	63	175	1.71	43	3	2,532	59.0	1,745	59.2	26	72.2	5.2	79.6	−24.4
J. G.	60	177	1.76	34	1	2,319	59.3	1,620	62.8	15	85.0	5.3	61.6	−31.7
N. C.	64	169	1.67	37	3	3,867	60.0	2,570	35.0	36	59.5	11.7	67.7	+22.6
E. S.	38	172	1.70	14	3	2,997	62.6	1,685	59.0	11	89.0	7.0	36.4	−12.1
W. H.†	64	170	1.98	35	2	2,842	62.8	1,700	57.5	40	62.0	8.3	80.0	−11.3
W. B.	47	176	1.73	26	1	3,988	64.3	2,220	48.8	35	67.5	10.0	71.5	+ 8.4
A. J.	59	186	1.78	43	3	2,931	65.0	1,575	56.5	21	78.8	6.9	67.6	−27.5
M. M.	65	158	1.59	55	1	2,561	65.0	1,380	61.0	27	66.4	7.7	70.4	−22.0
F. S.	41	175	1.74	21	3	3,260	65.2	1,740	60.0	16	86.0	7.5	57.0	−13.4
Av.	55	172	1.71	33		2,836	60.3	1,841	55.1	25	74.0	7.6	67.5	−14.1

* Diagnosis of tuberculosis made from examination of sputum on laboratories.

† Patient had cardiac disease with electrocardiographic evidence of myocardial damage.

sema was classified in five groups based on the residual air per cent of total lung volume as described below. The volume of residual air normally constitutes 25 per cent or less of the total lung volume. When such values are obtained the lungs as a whole are physiologically not emphysematous, although there may be small scattered or localized areas of bullae or blebs which are insignificant in their contribution to the pulmonary functional impairment by increasing the mixing and dilution factor. When the residual air occupies 25 to 35 per cent of the total lung volume a slight degree of emphysema is present, which again is not generally of sufficient magnitude to produce symptoms from the emphysema factor *per se*, that is, the mixing and dilution factor imposed by the increased volume of residual air. An occasional exception is represented by the patient with a marked decrease in vital capacity. If, however, the residual air is 35 to 45 per cent of the total lung volume, emphysema of a moderate degree exists. If the residual air is proportionately greater (45 to 55 per cent), the emphysema is advanced, and is far advanced when the residual air represents more than 55 per cent of total lung volume. In general, for the emphysema alone to alter pulmonary function significantly, the residual air must occupy 35 per cent or more of the total lung volume.

The data of the 100 cases are presented in table 1, and are arranged in five groups with respect to the severity of the emphysema. In each group the individual cases are listed in order according to the increasing percentage of total lung volume occupied by the residual air. The average values for each group are given in table 2, which is a summary of table 1.

The duration of exposure to dust inside the mines, as reported by the patient, ranged from 20 to 45 years, with the exception of four cases. There was no relationship between the length of exposure and the degree of pulmonary emphysema present (tables 1 and 2).

In the group of cases with a history of 20 to 25 years exposure (table 1), there were 25 patients. In 9 the degree of emphysema was advanced or far advanced, in 6 it was moderate and in 10, or 40 per cent, the emphysema was insignificant. The findings were similar in the group with a history of 35 to 40 years of inside exposure. Six of these 23 patients had an advanced or far advanced degree of emphysema, 6 had a moderate degree and 11, or 47.8 per cent, had insignificant emphysema. In this series one case with a history of 14 years of exposure had a far advanced degree of emphysema, but another case with 47 years of exposure had only a slight degree of emphysema. Two other patients with histories of exposure inside the mines of 55 and 56 years, respectively, had emphysema of a far advanced degree.

Figure 1 shows the relationship of age and residual air, expressed as per cent of total lung volume. It will be noted that 14 per cent of the cases had no emphysema and 23 per cent had only a slight degree of emphysema, making a total of 37 per cent without a significant degree of emphysema. However, 63 per cent of the cases did have a significant degree of emphysema distributed as follows: 30 moderate; 18 advanced; and 15 far advanced. Figure 1 reveals that the ages are, for the most part, between 40 and 65 years, and that all degrees of emphysema occur in all age groups. For example, of 37 in the 40 to 50 year age

group, a significant degree of emphysema was present in 59.4 per cent of the cases and an advanced degree in 32.4 per cent. In the 50 to 60 year age group, (38 cases) 63.2 per cent had a significant degree of emphysema and 28.9 per cent an advanced degree. In the group 60 years of age and above, (24 cases) 66.7 per cent had a significant degree of emphysema and 37.5 per cent an advanced degree.

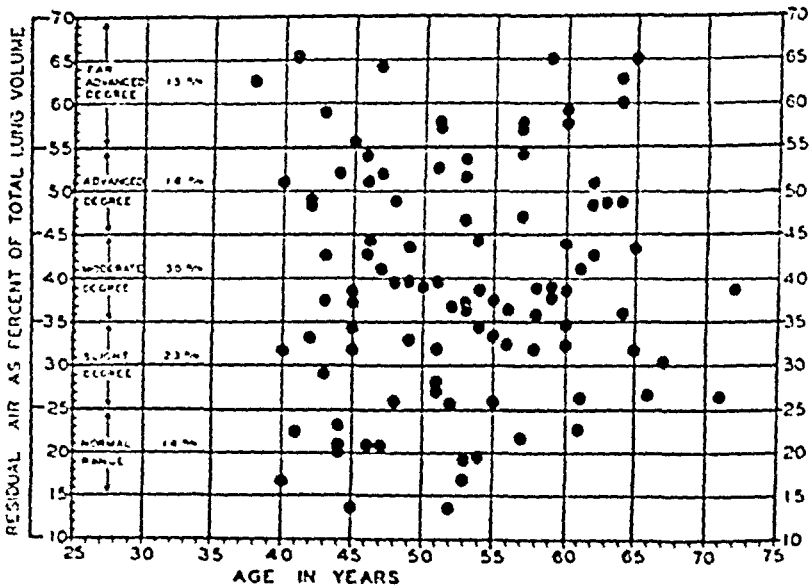


FIG. 1. Pulmonary emphysema in 100 cases of anthracosilicosis. Relationship between age in years and degree of pulmonary emphysema (as indicated by expressing the residual air as per cent of total lung volume on the ordinate) in 100 cases of anthracosilicosis. Each dot represents one patient. Note the occurrence of all degrees of emphysema in all age groups (40 to 65 years). In 14 per cent of the cases there was no emphysema and in 23 per cent only a slight degree of emphysema.

One patient, (ES table 1), 38 years of age with a 14 year history of exposure inside the mines had a far advanced degree of emphysema. It should be noted that 76 per cent of the patients studied were under 60 years of age and 31.6 per cent of these had an advanced and disabling degree of pulmonary emphysema.

Alveolar nitrogen: The alveolar nitrogen per cent after seven minutes of 99.6 per cent oxygen breathing was averaged by groups as shown below:

GROUPS	TOTAL NUMBER OF CASES	AVERAGE ALVEOLAR NITROGEN PER CENT	RANGE OF ALVEOLAR NITROGEN PER CENT
1. No emphysema	14	2.05	(1.00 to 4.22)
2. Slight emphysema	23	2.18	(0.81 to 4.38)
3. Moderate emphysema	30	3.40	(1.16 to 11.90)
4. Advanced emphysema	18	4.82	(1.41 to 9.65)
5. Far advanced emphysema	15	5.90	(2.67 to 11.80)

The average alveolar nitrogen per cent in the five groups shows a progressive increase as the degree of emphysema increases; however, the individual variations in each group are large.

Relationship between vital capacity and residual air: The relationship between vital capacity and the residual air expressed as per cent of total lung volume may be seen in figures 2 and 3. In general the decrease in vital capacity for the greater number of cases varied from 30 to 65 per cent below the predicted value. There

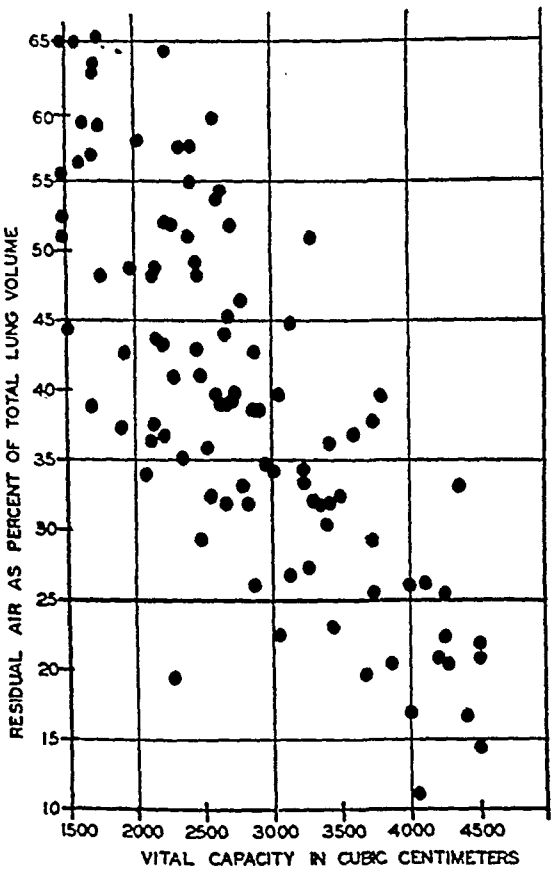


FIG. 2. Relationship between vital capacity and the degree of emphysema.

was poor correlation between vital capacity and the pulmonary emphysema present, the individual variations between cases with similar degrees of emphysema being great (figure 2). A vital capacity of 2,500 cc. may occur in the presence of no emphysema or in far advanced emphysema (figure 2). The average values for the groups classified by the degree of emphysema present (table 2 and figure 4), reveal a progressive decrease in the vital capacity as the residual air per cent of total lung volume increases.

The vital capacity (VC) data, given in table 1 and figure 3 may be further

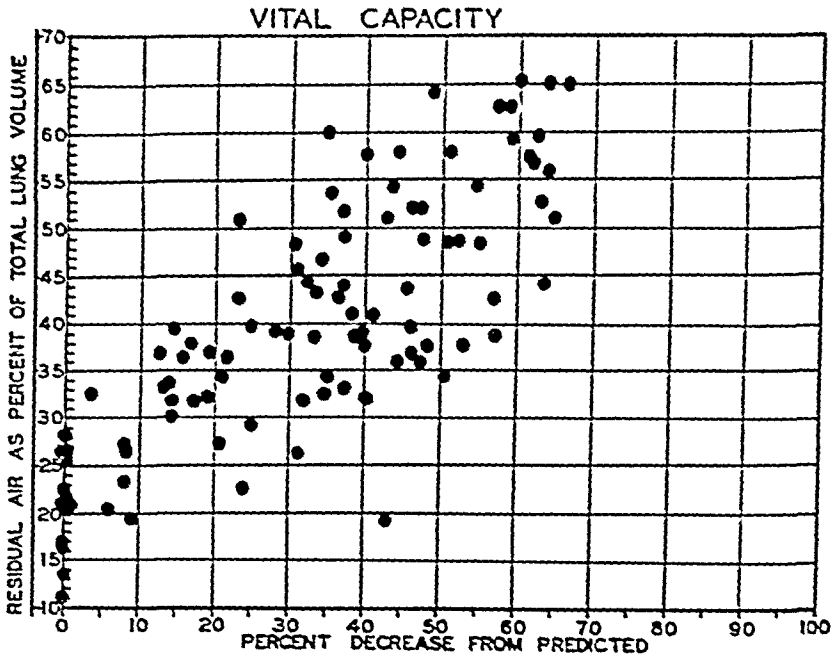


FIG. 3. Relationship between the per cent decrease in vital capacity from the predicted values and the degree of emphysema.

TABLE 2

Averages of values presented in table 1

Group	Number Cases	Age	Height	Body Surface Area	Exposure Inside Mines	Residual Air		Vital Capacity		Maximal Breathing Capacity		Ventilation	Breathing Reserve of Maximal Breathing Capacity	Total Lung Volume Change from Predicted
						Observed	Total Lung Volume	Observed	Decrease From Predicted	Observed	Decrease From Predicted			
		Yrs.	cm.	sq. m.	Yrs.	cc.	per cent	cc.	per cent	L. per minute	per cent	L./minute	per cent	per cent
I	14	49	171	1.82	28	936	19.2	3,910	6.5	83	26.1	8.0	89.0	−10.0
II	23	54	170	1.78	31	1,420	30.8	3,270	19.1	66	36.3	9.0	85.0	−11.5
III	30	54	170	1.73	32	1,703	39.6	2,637	35.8	46	55.1	8.4	79.6	−20.1
IV	18	52	171	1.73	29	2,305	50.4	2,315	43.3	34	67.0	8.2	73.8	−15.2
V	15	55	172	1.71	35	2,836	60.3	1,844	55.1	25	74.0	7.6	67.5	−14.4

Group I. Normal or no pulmonary emphysema.
Group II. Slight degree of emphysema.
Group III. Moderate degree of emphysema.
Group IV. Advanced degree of emphysema.
Group V. Far advanced degree of emphysema.

grouped for analytical study based on the decrease from the predicted values as follows:

(1) Degree of emphysema not significant, 37 cases.
  19 cases (51.4 per cent).....Decrease in VC 10 per cent or less
  10 cases (27.0 per cent).....Decrease in VC 10 to 30 per cent
  8 cases (21.6 per cent).....Decrease in VC 30 to 50 per cent

(2) Degree of emphysema significant, 63 cases.
  10 cases (15.9 per cent).....Decrease in VC 10 to 30 per cent
  32 cases (50.8 per cent).....Decrease in VC 30 to 50 per cent
  11 cases (17.4 per cent).....Decrease in VC 50 to 60 per cent
  10 cases (15.9 per cent).....Decrease in VC 60 to 70 per cent

(3) In the group of 33 patients with an advanced and disabling degree of emphysema, 16 (48.5 per cent) had a vital capacity which was decreased less than 50 per cent from the predicted value.

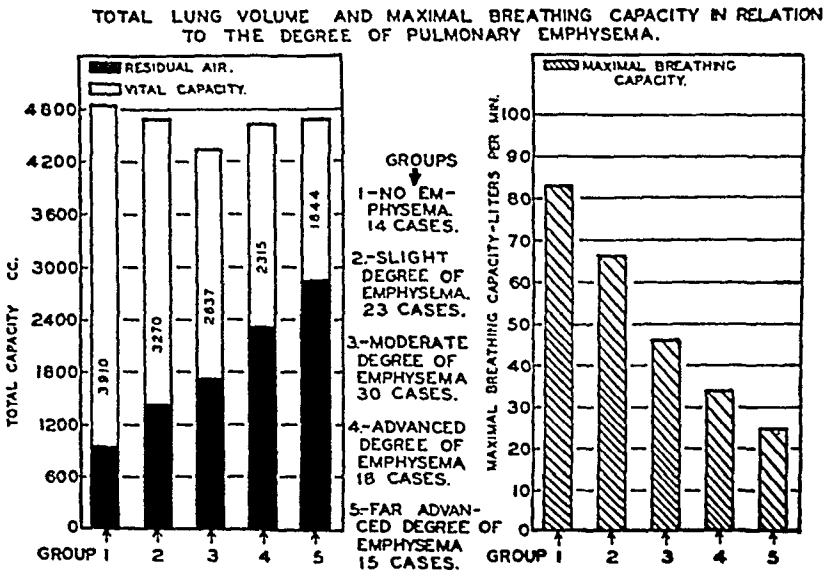


FIG. 4. Total lung volume and maximal breathing capacity in relation to the degree of pulmonary emphysema.

Maximal breathing capacity: The maximal breathing capacity (MBC) was measured at the same time as the vital capacity. The relationship between MBC and the degree of pulmonary emphysema is presented in figures 5 and 6. There is a better correlation between the degree of pulmonary emphysema present and the reduction in MBC observed than was found to exist between vital capacity and emphysema. In 56 cases the MBC was reduced 50 to 80 per cent from the predicted value and in 25 cases the decrease was 70 per cent or more from the predicted value (figure 6). The decrease in MBC from the predicted value, as shown in figure 6 and table 1, may be similarly grouped for analysis as was the vital capacity:

(1) Degree of emphysema not significant, 37 cases.
  5 cases (13.5 per cent).....Decrease in MBC 10 per cent or less
  11 cases (29.8 per cent).....Decrease in MBC 10 to 30 per cent

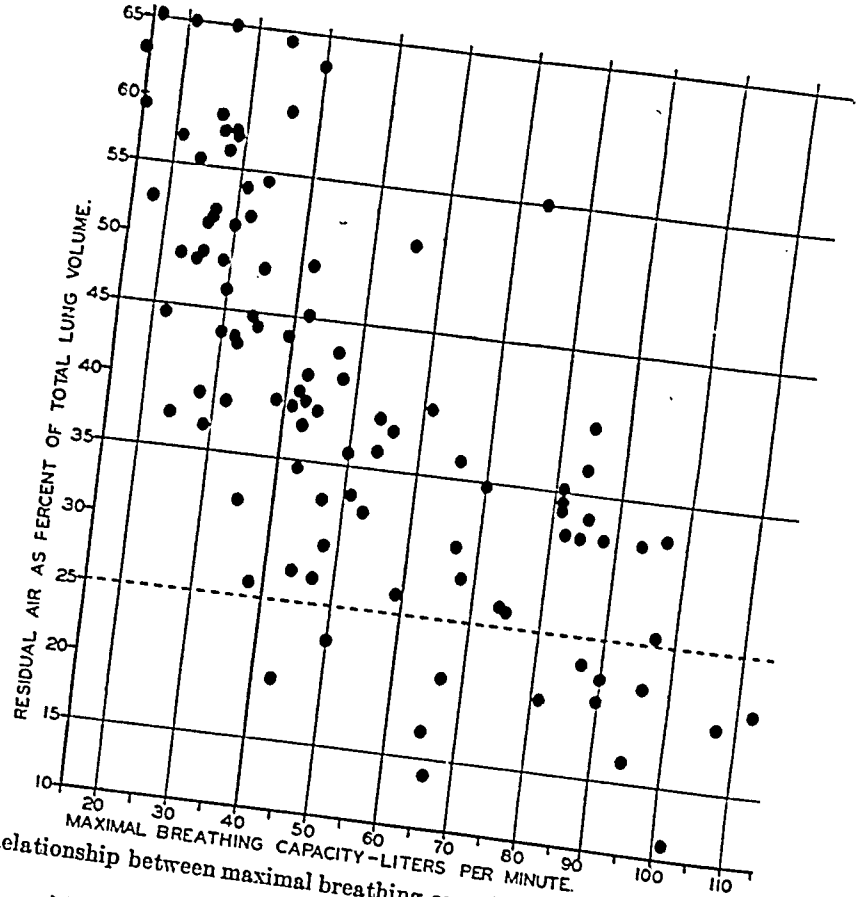


FIG. 5. Relationship between maximal breathing capacity and the degree of emphysema.

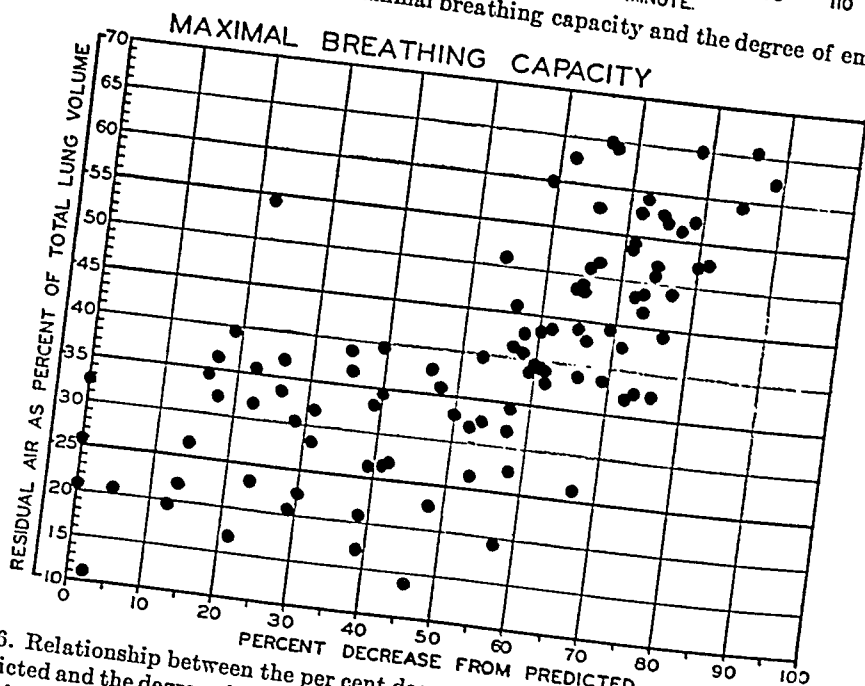


FIG. 6. Relationship between the per cent decrease in maximal breathing capacity from the predicted and the degree of emphysema. Note the greater decrease as compared to vital capacity in figure 3.

12 cases (32.4 per cent).....Decrease in MBC 30 to 50 per cent
8 cases (21.6 per cent).....Decrease in MBC 50 to 60 per cent
1 case (2.7 per cent).....Decrease in MBC 60 to 70 per cent

(2) Degree of emphysema significant, 63 cases.
5 cases (7.9 per cent).....Decrease in MBC 10 to 30 per cent
5 cases (7.9 per cent).....Decrease in MBC 30 to 50 per cent
9 cases (14.3 per cent).....Decrease in MBC 50 to 60 per cent
19 cases (30.3 per cent).....Decrease in MBC 60 to 70 per cent
20 cases (31.7 per cent).....Decrease in MBC 70 to 80 per cent
5 cases (7.9 per cent).....Decrease in MBC 80 to 90 per cent

(3) In the group of 33 patients with an advanced and disabling degree of pulmonary emphysema, all except one had 50 per cent or greater decrease in MBC.

In general, MBC shows a much better correlation with the disability produced by the pulmonary emphysema than is shown by the vital capacity. Although in some instances the variations in individual cases with the same degree of emphysema are large, they are not as great as are seen in the vital capacity measurements (figures 2 and 5). The average for the five groups (table 2, figure 6), shows a smooth progressive decrease in MBC as the residual air increases and the vital capacity decreases. If the MBC is decreased 60 per cent or more from the predicted value or if the actual measurement for men is 40 L. per minute, or less, the degree of emphysema present is usually significant (figures 5 and 6).

Hyperventilation: Hyperventilation was present in all groups (tables 1 and 2). A few patients in each group exhibited marked hyperventilation, in some instances resulting from apprehension and in others as a compensatory response to the emphysema. Although the resting minute ventilation has limited prognostic value as a single measurement, it is useful in determining the breathing reserve. The breathing reserve expressed as per cent of MBC was 89.6, 85.0, 79.6, 73.8 and 67.5 respectively for the five groups (tables 1 and 2). The breathing reserve when expressed in this manner shows a fair correlation with the degree of emphysema present (figure 7). If the breathing reserve was 70 per cent or less of the MBC, the degree of emphysema was significant but was insignificant if the breathing reserve was 90 per cent or more of the MBC.

The total lung volume was decreased in 81 out of 100 cases. The most marked decrease, an average of 20 per cent, was in the third group with a moderate degree of emphysema (figure 4 and table 2). In this group a proportionally greater decrease in vital capacity and smaller increase in residual air were present as compared to the other groups. Patients with an advanced degree of pulmonary emphysema apparently have developed more compensatory emphysema with a resulting smaller decrease in total lung volume. Why some patients develop so much more compensatory emphysema than others is not known. The extent of the emphysema in this group at least was not apparently related to age, height, weight, or length of exposure in mines.

Roentgenographic findings: The chest roentgenograms were classified according to the descriptions of Pancoast and Pendergrass (7) (table 1). In this classification the first stage is characterized by increase in the size and density of the hilar

lymph nodes, perivascular thickening outward toward the pleura, and a slight fine mottling in the parenchyma; the second stage by typical nodular round and

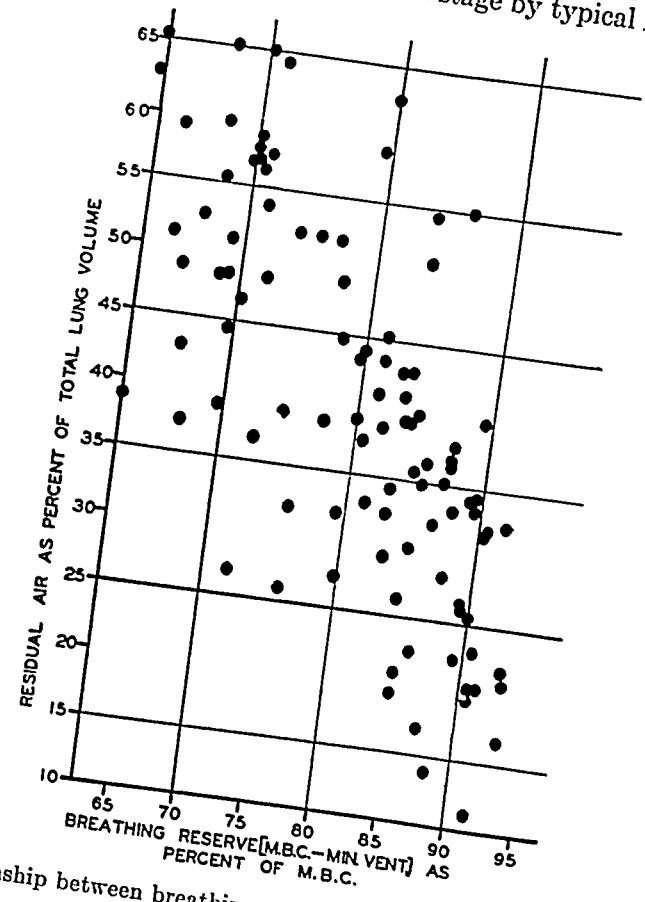


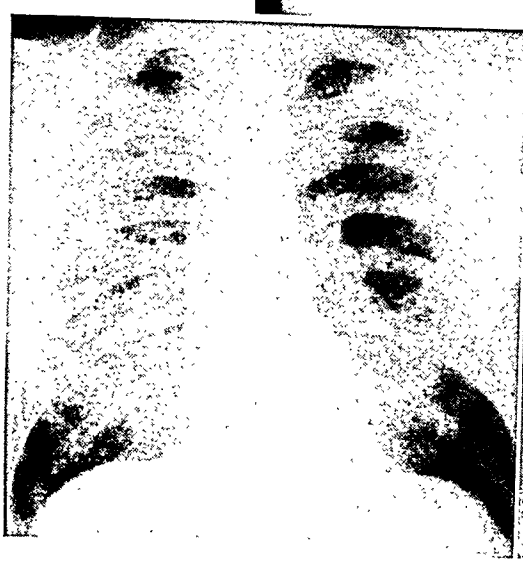
FIG. 7. Relationship between breathing reserve and the degree of emphysema.

TABLE 3

Stage of silicosis based on roentgenogram, and degree of pulmonary emphysema as determined from measurements of residual air and total lung volume

DEGREE OF EMPHYSEMA	TOTAL NUMBER OF CASES	FIRST STAGE	SECOND STAGE	THIRD STAGE
1. None	14	1	5	8
2. Slight	23	4	4	15
3. Moderate	30	3	6	21
4. Advanced	18	0	3	15
5. Far advanced	15	4	3	8

oblong shadows of soft even density from 2 to 6 mm. in diameter radiating from the hilus; and the third stage by the presence of shadows larger than 6 mm. in diameter, more numerous and with coalescence into aggregates. In this series of 100 cases, 67 were classified as third stage, 21 as second stage, and 12 as first



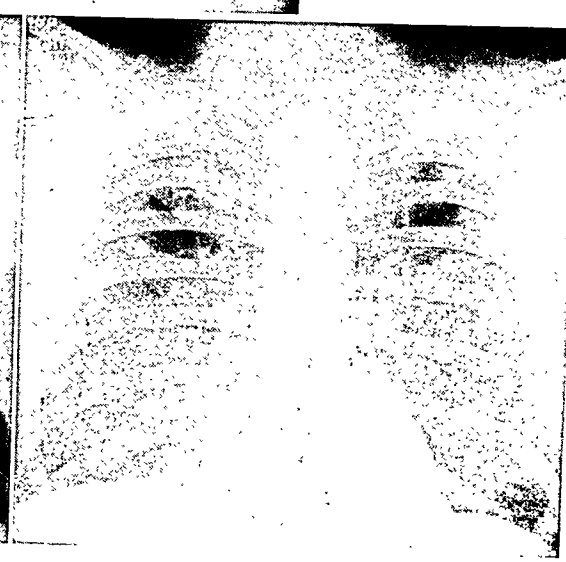


FIG. 8. (Upper) Typical roentgenogram of the chest taken at full inspiration in a case with far advanced emphysema. History of 43 years inside mine work. Present complaints: marked dyspnea and weakness, slight cough and expectoration and 55 lb. weight loss. Age 63. Residual air 2,532 cc., vital capacity 1,745 cc., residual air equals 59 per cent of total lung volume, and maximal breathing capacity 26 L. per minute. Good correlation between roentgenographic findings and studies of pulmonary function.

FIG. 9. (Lower left) A case illustrating poor correlation between roentgenographic findings (inspiratory film) and studies of pulmonary function. History of 20 years inside mine work. Present complaints: dyspnea on effort, cough and expectoration, weakness and 15 lb. weight loss. Age 41. Residual air 1,219 cc., vital capacity, 4,250 cc., residual air equals 22.3 per cent of total lung volume, and maximal breathing capacity 90 L. per minute. No emphysema present.

FIG. 10. (lower right) Another example showing discrepancy between roentgenographic findings (inspiratory film) and the studies of pulmonary function. History of 28 years inside mine work. Present complaints: dyspnea, marked cough and expectoration and upper chest pain. Age 48. Residual air 2,503 cc., vital capacity 3,800 cc., residual air equals 39.6 per cent of total lung volume, maximal breathing capacity 61 L. per minute. Moderate degree of emphysema present.

stage. In a few of the cases classified as first stage silicosis there was question as to whether or not the degree of exaggeration of the lung marking was within normal limits. Four of the 12 cases with first stage silicosis had a far advanced degree of emphysema. There was no apparent relationship between the stage of silicosis, as estimated from roentgenograms, and the degree of pulmonary emphysema (table 3).

The 67 cases of third stage silicosis present two points of interest: (*1*) the lack of correlation between the roentgenographic findings and the emphysema; and (*2*) the distribution relationship between the emphysema and the total number of cases in each group. In the cases with a far advanced degree of emphysema and third stage silicosis, the chest roentgenogram was satisfactory for predicting the degree of emphysema and the limitation in maximal breathing capacity (figure 8). However, in other cases a wide discrepancy was observed (figures 9 and 10). The movement of the diaphragm on fluoroscopy and as seen on inspiratory and expiratory films and changes in illumination of the lungs between inspiration and expiration were most helpful in predicting the degree of emphysema present. In many cases, however, the predicted values were erroneous.

DISCUSSION

In 100 anthracite coal miners the maximal breathing capacity, vital capacity, minute ventilation, breathing reserve, duration of exposure in mines, and the roentgenographic changes have been correlated with the degree of pulmonary emphysema. The maximal breathing capacity (MBC) was observed to show the best single correlation. If the maximal breathing capacity was less than 40 L. per minute, the emphysema present was a significant factor in the impairment of pulmonary function. Vital capacity as a single measurement was of limited value in predicting the extent of the emphysema, and could not be relied upon as a quantitative measure of pulmonary functional impairment. In the present study the greatest value of vital capacity determinations was in the calculation of total capacity. The decrease in vital capacity below the predicted value was consistently less than was the reduction in maximal breathing capacity, a relationship also observed by others (8). No constant relationship exists between vital capacity and maximal breathing capacity, however, and, with few exceptions, the MBC measurements were a more reliable indication of the degree of disability than were the deviations in vital capacity. In the present study maximal breathing capacity was the best single test of the ventilation factor of pulmonary function. This results from the facts that MBC is concerned with the movement of the diaphragm, the intercostals, and other accessory muscles of respiration, the patency of the tracheobronchial pulmonary airway, the elastic property of lung tissue, and neuromuscular coordination. Maximal breathing capacity measurements are simple to perform and offer a practicable test of pulmonary function for mass examinations. If MBC measurements are used routinely along with the chest roentgenogram, the efficiency of the search for early evidence of silicosis should be greatly improved.

There was no demonstrable relation between exposure time in the mines and the degree of emphysema present. The findings in the present study suggest

that individual susceptibility to exposure of coal mine dust varies and unknown intrinsic factors may exert an influence on the degree of pulmonary disability that follows. Bamberger (9) has reported that, in a group of miners working under the same conditions at the same time, one may develop linear exaggeration of a mild degree, whereas another will develop far advanced nodular silicosis. The condition of the ciliated epithelium (10) may be responsible for the lack of a normal degree of protection against dust invasion. Decreased motility of the cilia in certain persons impairs the removal of inhaled particles. The exposure time as given by the patient has a limited value, although in the present study as exact a history as possible of the total time inside the mines was sought. As many accurate measurements of the degree of emphysema have been made, the observations reported in table 1 suggest the presence of an individual susceptibility factor. For example, case F. R., age 54 (group 1, table 1), with a 40 year history inside the mines had no emphysema, while case F. S., age 41 (group 5), with a 21 year history inside the mines had a far advanced degree of emphysema. Although the concentration of dust in the mines varies widely, it is unlikely, even if the exact amount of dust inhaled could be ascertained, that it could be correlated with the degree of emphysema present in this series of 100 cases.

The findings indicate that practically all of the subjects hyperventilated, but there was no significant correlation between the extent of hyperventilation and the degree of emphysema. Hyperventilation is a compensatory mechanism, however, and during the early stages of the process may be adequate until loss of elasticity of the lungs and obstruction markedly decrease MBC and VC. The minute ventilation alone was not related to the extent of pulmonary disability. In most cases the total lung volume was decreased. Nineteen per cent of the cases showed an increase, however, and in a few there was no significant change. Compensatory emphysema was present in some cases and absent in others. Some of the patients with flat chests had an advanced degree of emphysema, probably indicating a more rapid development of the emphysema and the onset of the process later in life than in the case of the typical "barrel" shaped chest of the asthmatic with a far advanced degree of emphysema.

The roentgenographic findings alone do not permit a proper evaluation of the impairment of pulmonary function in many cases. Extensive deposits of silica may be demonstrated on the roentgenogram when the pulmonary function is only moderately altered, while in other cases with minimal findings on the roentgenogram, the pulmonary function is greatly impaired. In a considerable number of cases even the degree of emphysema present is difficult to approximate from the roentgenogram alone.

As 14 per cent of the cases in this series had a residual air less than the normal predicted, and 23 per cent had only a slight increase in the residual air per cent of total lung volume, pulmonary emphysema could not be assigned as the basis of the respiratory difficulty. Furthermore, in most instances those subjects with an insignificant amount of emphysema did not have a reduction in MBC of sufficient degree to account for the symptoms presented.

At the present time the studies are being extended to investigate the nature of different types of impairment of pulmonary function including the transfer

of O_2 and CO_2 between the alveoli and arterial blood. Preliminary findings indicate that the fibrosis present in some cases of anthracosilicosis, although not producing a significant degree of emphysema, interferes with the uniform aeration of the alveoli (the distribution factor (11, 12)). Consequently an increased percentage of alveoli are poorly ventilated with a resulting increase in the mean pressure gradient of pO_2 between the alveoli and the arterial blood. Distribution, one of the factors contributing to the alveolar-arterial pO_2 gradient, may be defined as: (*1*) the variations in alveolar pO_2 in different parts of the lungs; and (*2*) the ratio of alveolar ventilation to alveolar perfusion. Also the fibrosis obliterates the blood supply to alveoli, so that areas of the lung are ventilated but not perfused, thus increasing the physiological dead space and reducing the efficiency of O_2 removal per liter of ventilation. Furthermore, studies with high and low levels of oxygenation indicate that the increased pO_2 gradient between the alveoli and arterial blood is primarily one of distribution and not a diffusion difficulty (increased resistance to passage of oxygen through the pulmonary membrane). The pulmonary functional impairment of distribution described above may produce marked arterial oxygen unsaturation at rest even in the absence of emphysema and with a normal maximal breathing capacity. An accurate clinical evaluation of the disability in such a patient is very unlikely when the roentgenographic findings are minimal.

The compensatory mechanism for the mixing and dilution problem imposed by the increased volume of residual air is hyperventilation. The extent of this response varies widely, so that compensation is good in some patients and poor in others with the same amount of emphysema. Fibrosis and loss of elasticity of the chest wall structures and lungs restricts the amount of hyperventilation possible without dyspnea. The dyspnea level varies widely, a fact which explains the observations that the severity of respiratory symptoms are not an accurate measure of the relationship between the residual air and the total lung volume.

SUMMARY

The degree of pulmonary emphysema has been evaluated physiologically, and ventilation measurements obtained, in 100 unselected cases of anthracosilicosis with pulmonary symptoms and a long history of exposure to mine dust.

A quantitative indication of the degree of emphysema was obtained by expressing the residual air as per cent of the total lung volume. On the basis of this value all cases were classified in five groups as follows: (*1*) no emphysema (25 per cent or less); (*2*) slight degree of emphysema (25 to 35 per cent); (*3*) moderate degree of emphysema (35 to 45 per cent); (*4*) advanced degree of emphysema (45 to 55 per cent); and (*5*) far advanced degree of emphysema (65 per cent or more).

No relationship existed between the history of length of exposure to mine dust and the degree of pulmonary emphysema present. Moreover, there was no apparent relationship between the stage of silicosis, as determined from the roentgenogram, and the degree of pulmonary emphysema present. Most of

the subjects were between 40 and 65 years of age and all degrees of emphysema were present in all age groups.

The degree of emphysema present was better correlated with the maximal breathing capacity than with the vital capacity. The decrease from the predicted value was consistently greater for maximal breathing capacity than vital capacity. If the maximal breathing capacity was below 40 L. per minute or decreased 60 per cent or more from the predicted, the amount of emphysema present was significant in producing impairment of pulmonary function.

The routine use of maximal breathing capacity measurements along with the chest roentgenogram should enhance the efficiency of the search for early evidence of silicosis.

Measurements of resting minute ventilation revealed that hyperventilation was consistently present. The average values for breathing reserve in the different groups show a progressive decrease as the degree of emphysema increases although wide individual variations exist.

Pulmonary emphysema accounts for part of the impairment of pulmonary function in anthracosilicosis. In a considerable number of cases, however, other factors must be responsible because the amount of emphysema present is not significant.

SUMARIO

Enfisema Pulmonary Mediciones de la Ventilación en Cien Mineros de Carbón de Antracita con Síntomas del Aparato Respiratorio

En 100 casos no seleccionados de antracosilicosis con síntomas pulmonares y prolongada historia de exposición al polvo de minas, se valuó fisiológicamente la intensidad del enfisema pulmonar y se obtuvieron mediciones de la ventilación.

Expresando el aire residual en por ciento del volumen pulmonar total, se obtuvo una indicación cuantitativa del grado del enfisema. A base de esa cifra, todos los casos fueron clasificados en cinco grupos, a saber: (1) sin enfisema (25 por ciento o menos); (2) enfisema leve (25 a 35 por ciento); (3) enfisema moderado (35 a 45 por ciento); enfisema avanzado (45 a 55 por ciento), y enfisema muy avanzado (65 por ciento o más).

No hubo relación entre la historia de la duración de la exposición al polvo de mina y la intensidad del enfisema pulmonar presente. Tampoco existía aparentemente entre el período de la silicosis, determinado por la radiografía, y la intensidad del enfisema pulmonar. La mayor parte de los sujetos tenían de 40 a 65 años de edad, observándose en todos los grupos etarios los diversos grados del enfisema.

La intensidad del enfisema presente se correlacionó con la capacidad respiratoria máxima mejor que con la capacidad vital, siedno la disminución de la cifra anticipada constantemente mayor para la primera que para la segunda. Si la capacidad respiratoria maxima era inferior a 40 L. por minuto o quedaba 60 por ciento o más por debajo de lo anticipado, la intensidad del enfisema presente afectaba significativamente la función pulmonar.

El empleo sistemático de las mediciones de la capacidad respiratoria máxima,

junto con la radiografía torácica, debe acrecentar la eficacia de la pesquisa de signos temperanos de silicosis.

Las mediciones de la ventilación por minuto en reposo revelaron constantemente la presencia de hiperventilación. Las cifras medias de la reserva respiratoria en los diversos grupos muestran, aunque existen variaciones individuales, una disminución gradual a medida que se acentúa el enfisema.

El enfisema pulmonar explica parte de la reducción de la función pulmonar en la antracosilicosis, pero en una considerable proporción de casos, deben intervenir otros factores por no revestir importancia el enfisema presente.

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MANAGEMENT OF TUBERCULOSIS IN PSYCHOTIC PATIENTS[1, 2]

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INTRODUCTION

The purpose of this paper is to instill optimism and to eliminate some of the pessimism that has often pervaded the literature regarding the problems of tuberculosis that exist in most large mental institutions. The fact that an appreciable number of psychotics make at least a partial recovery and return to their families and home communities and the problem of control of the disease in the institution make a more active approach desirable.

There have been many articles in reference to tuberculosis among the mentally ill (1, 2, 3, 4). However, reference to the literature has not been successful in yielding much information in regard to treating these patients. In the majority of instances an attitude of pessimism has prevailed. This is particularly true in regard to collapse therapy and thoracic surgery. For example, Wicks (5) states "it is rather doubtful whether any of the more radical types of chest surgery (with the possible exception of intrapleural pneumonolysis) should be attempted in a tuberculosis mental unit." Leonidoff (6) states "pneumothorax is unsuitable for the mentally ill patient". Although it is not intended to minimize the difficulties involved in treating psychotic patients with pulmonary tuberculosis, the writers believe that a more optimistic attitude is warranted. To support that contention is the purpose of the present report which is based on experiences with 100 cases of pulmonary tuberculosis treated at the Veterans Administration Hospital, North Little Rock, Arkansas, during the past two years. Although the period of observation has been of insufficient length to permit statistically valid conclusions as to the final result of treatment, it is believed that these experiences show that the problems in the management of psychotic tuberculous patients are not insurmountable.

Detection of Cases

The psychotic tuberculous patient probably represents a great source of infection and propagation of the disease, both in patient population and in their home communities. Bogen, Tietz and Grace (7) in 1934 surveyed the patients in Longview State Hospital, Cincinnati, Ohio. Their survey showed that tuberculous infection among mental hospital patients increases according to length of their stay in institutions and indicated that probably the majority of such patients contract their infection as well as their disease after their admission to the mental hospital. The present survey has revealed 28 patients who apparently

[1] From the Medical Service, Veterans Administration Hospital, North Little Rock, Arkansas.

[2] Published with permission of the Chief Medical Director, Department of Medicine and Surgery, Veterans Administration, who assumes no responsibility for the opinions expressed or conclusions drawn by the authors.

contracted their infection in the hospital. The danger of spread of tuberculosis in home communities arises from: (*1*) discharge of patients on trial visit[3]; and (*2*) elopement of patients who have unrecognized active pulmonary tuberculosis. The present survey has revealed that 5 patients now under care were discharged in former years at a time when they probably had active pulmonary tuberculosis and thus undoubtedly were in a position to spread the disease.

How can these cases be discovered? The problem is difficult enough in mentally competent patients for it is well recognized that the tubercle bacillus may produce disease without early prominent symptomatology. It is even more difficult to detect the disease among psychotic patients. The latter often do not complain and many times when they do their complaints are quite bizarre. Too much dependence cannot be placed on physical signs for often the psychotic patient will not cooperate sufficiently to permit proper auscultation of the chest. Weight loss is a common finding in many psychotic patients and certainly it is not a specific sign of any one disease. Low grade fever may often be overlooked unless the temperature is taken at frequent intervals, a practice which is not usually a routine on most psychotic wards. This problem of detection of tuberculosis in psychotic patients has previously been well studied at the Ontario Mental Hospitals (Canada) (1). Reference will be made to these studies in the subsequent discussion.

Tuberculin testing is obviously not the answer. McGhie and Brink (8) tested 1,209 patients with tuberculin and 91.5 per cent had positive reaction whereas only 5 per cent had significant pulmonary lesions on roentgenography. The Ontario Mental Hospital group first used fever as a method of screening. The axillary temperatures of all patients were determined during one week out of every four months at 4 p.m. or 8 p.m. All patients with temperatures above 98° F. for four days or more were investigated for possible pulmonary tuberculosis. After the temperature observations, the erythrocyte sedimentation rates were determined for all patients; those with rates greater than 10 per cent in the first hour (Westergren Method) were investigated for possible pulmonary lesions. Roentgenographic screening was performed after temperature surveys had been conducted every four months for two years, and six months after a blood sedimentation rate survey. The roentgenographic examinations revealed that 2 per cent of the patients, unsuspected of disease on the basis of both temperature and blood sedimentation rate determinations, had lesions which were later proved to be active pulmonary tuberculosis. An additional 2.3 per cent of the patients had lesions the activity of which was questionable.

In July, 1946 there were 29 patients with active disease hospitalized on the tuberculosis ward at the Veterans Administration Hospital, North Little Rock, Arkansas. The patient population of the hospital at that time averaged between 1,600 and 1,800 patients. Roentgenographic surveys were begun. Since that time 76 patients with evidence of active pulmonary tuberculosis have been

[3] A probationary ninety day period during which time the patient is released in the custody of his nearest relative. If his adjustment is not satisfactory, he is returned to the hospital; otherwise, he is discharged and resumes his place in the community.

In each case treatment is outlined as would be prescribed for a mentally well person with tuberculosis of comparable severity. Treatment is then modified as dictated by the psychiatric condition.

Bed-rest: It is impossible to obtain as strict bed-rest as can be obtained in mentally competent patients, with one exception. The exception is provided by the patients whose illness has marked catatonic features. Nevertheless, a modified rest regimen is possible. If the patient is permitted to sit in a chair near his bed for a few specified hours per day, it often results in his accepting much more of a limitation of activity then he would observe with rigidly enforced bed-rest. In the majority of cases it has been possible to keep patients in bed for at least eighteen hours each day. This accomplishment is largely a result of the attitude of the nurses and attendants. Constant persuasion must be combined with friendliness and understanding. Restraint should never be used to try to maintain rest therapy though it sometimes becomes necessary in management of the patient's psychosis.

Once a routine of rest in bed with toilet privileges and specified hours of limited activity has been established, most of the newly admitted patients will follow this example to a surprising degree. In the presence of hyperactivity, moderate sedation should be added to the program. Phenobarbital in dosages varying from 0.03 Gm. to 0.09 Gm. three times daily has been very helpful.

The problem is best exemplified by the following case history of a difficult problem in management.

The patient, H. T., was a 24 year old white single male. In September, 1944 he became frankly psychotic and was hospitalized at the Veterans Hospital, North Little Rock, on October 5, 1944. A diagnosis of schizophrenic reaction, catatonic type, was made and he received twenty-five electric shock treatments. He showed some improvement and became able to care for himself but remained emotionally blunted, manneristic with signs of deterioration. Roentgenographic examination of the chest upon admission in October, 1944 revealed no evidence of tuberculosis. Chest roentgenogram in April, 1946 was reported as negative but in retrospect a small infiltration was present in the left lung. Roentgenogram of the chest in June, 1947 revealed an infiltration involving two-thirds of the left lung. Gastric lavage examination was positive for acid-fast bacilli on two occasions. A diagnosis of pulmonary tuberculosis, moderately advanced and active was made. Upon first being hospitalized on the tuberculosis service, the patient ate extremely poorly and was very hyperactive and refused to remain in bed. He did not resist being put to bed but refused to stay there. He continually asked to be put on an outside work detail. The attendants were instructed to check his bed frequently and to find the patient and put him back to bed whenever he was found to be up. He was told repeatedly why it was important that he remain in bed. He showed no real insight concerning this matter. An attendant was specially detailed to observe the patient during meal time and feed him, if necessary. He was given phenobarbital 0.09 Gm. three times daily. The sedation caused the patient to be less active. After approximately one month of constant attention, patient began observing bed-rest adequately. It was finally discovered that the patient could be induced to eat by refusing him coffee until he had eaten all of his meal. There was a spread of his disease to the opposite lung soon after he entered the tuberculosis service. However, a bilateral pneumothorax has been instituted successfully, and the patient now observes fairly good ward routine. His prognosis is believed to be good in regard to his pulmonary disease.

Although the above principles of therapy were formulated independently, it was later found that the conclusions were the same as those of the group at the Ontario Mental Hospital.

Nutrition: The problem of nutrition is the second problem of importance in each patient. The dietary regimen followed is the same as in the hospitals for the mentally competent. However, the coexistence of psychosis often interferes with adequate nutrition. The patient with a paranoid personality may harbor the delusion that the food is poisoned, the catatonic patient, in his retreat from reality, will cease to eat, and the hebephrenic patient may play with his food rather than eat it. Those who present eating problems must be picked out and given special attention. Every possible solution should be given a trial. The same attitude of constant persuasion combined with friendliness is essential. Some must be fed as a child is fed. Some will eat when fed by a woman but will refuse all food offered by a man. Only in a few paranoic patients and in those with the most severe catatonia does gastric lavage become necessary.

The following case represents a typical example of a feeding problem.

The patient was a 56 year old white male who was admitted to the Veterans Administration Hospital, North Little Rock, December 19, 1924. A diagnosis of schizophrenic reaction, catatonic type, was made. The patient has been a feeding problem throughout his period of hospitalization. He is apathetic, mute, negativistic and at times resistive. A progress note in 1930 notes the following: "The patient still will eat for only one attendant and when that attendant is off duty, refuses to eat. He will take exercise only under the supervision of that one attendant." The patient was given eight electric shock treatments in 1946 but this did not improve his condition.

Roentgenographic examinations of the patient's chest were negative until 1946. In August, 1946 he was noted to have developed infiltration and cavitation in the left lung. Four sputum examinations were positive for acid-fast bacilli. The patient's psychiatric condition is such that he is not a candidate for major thoracic surgery. Minor collapse measures have failed. During his period of hospitalization on the tuberculosis service he has continued to present a problem in nutrition. At intervals he refuses to eat altogether. After being tube fed for periods varying from one to five weeks, he will suddenly resume eating voluntarily. He consistently refuses to eat from a tray but will eat in the dining hall at times. He eats better when fed by a woman than by a man. He often will take liquid nourishment and steadfastly refuse solid food.

Laboratory Investigations: The problem of laboratory investigation of the etiology and activity of the pulmonary lesions is also in need of special attention. Isolation of *M. tuberculosis* is necessary to establish the diagnosis. This presents many problems not encountered in the mentally competent patient. Sputum specimens are often difficult and sometimes impossible to obtain. Methods must be utilized to prevent one patient from using another's sputum cup. If all efforts to obtain sputum specimens from a patient are unsuccessful, gastric lavage must be performed. The initial use of this procedure often necessitates active restraint of the patient. It is significant, however, that most psychotic patients soon learn to undergo lavage without objection. The lavage specimens may be examined by direct smear but positive reports should be verified by culture or animal inocculation.

Roentgenographic evidence of cavitation and progression or regression of pulmonary infiltration in serial films is unquestionable proof of activity. However, lack of change does not necessarily mean that a particular lesion is arrested. Moreover, the etiology of a pulmonary lesion cannot be definitely established merely by its appearance on a roentgenogram.

Additional measures, such as an elevated erythrocyte sedimentation rate or monocytosis, as opposed to lymphocytosis, in the peripheral blood, are suggestive of activity but are nonspecific. The same is true of such clinical findings as fever and weight loss. A negative skin tuberculin reaction is believed to be strong evidence against the presence of tuberculosis in all except miliary or terminal pulmonary infections.

Laboratory investigations have been stressed because this problem often seems insurmountable to physicians unaccustomed to working with psychotic patients. The importance of the problem of differentiating arrested and active lesions is self-evident. In the writers' opinion, the conservative attitude of the group working at the Ontario Mental Hospital is to be commended. They recommended that patients who had once had proved active pulmonary tuberculosis should have negative sputum examinations for tubercle bacilli, normal erythrocyte sedimentation rates, and stationary lesions, as determined by serial roentgenography, for one year before being returned to contact with nontuberculous patients. During the latter six months of this period, the patient should be allowed full exercise and light work, if his mental condition permits. The Canadian investigators believed that it is extremely doubtful whether patients with moderately advanced or far advanced pulmonary tuberculosis should ever be returned to contact with nontuberculous patients because of the great possibility of relapse accompanied by a return of infectiousness. Burns (12) reiterates this philosophy when he states, "In our cases, those becoming chronic with stationary stabilized infiltrations tend to become more and more self-limited and, with the increasing age of the lesion, of little danger to the future well-being of the host. Only as they remain carriers of the tubercle bacilli do they play an important part in our tuberculosis control program."

Active Therapy

Thoracoplasty, pneumothorax, pneumoperitoneum, and phrenico-exeresis have all been utilized in the treatment of the patients in the present series. Closed intrapleural pneumonolysis has been employed when indicated. The criteria for the selection of cases for collapse therapy, in reference to the type and status of the patient's pulmonary lesion, are the same as those used in mentally competent patients. The early institution of collapse therapy is favored, however, because: (*a*) many psychotic patients are unable to follow the bed-rest schedule and it is felt that such patients should be given the benefit of collapse therapy at an early date; (*b*) those psychotic patients who will follow a bed-rest schedule, such as those with well developed catatonic features, are just the ones whose mental condition would be benefitted by encouraging them to indulge in more activity. Thus, the sooner their pulmonary lesion is in a state to allow activity

the quicker the more active measures can be taken in treating their neuropsychiatric condition.

The psychiatric status must also be taken into consideration in each individual case. In general, it is felt that collapse therapy, including major thoracic surgery, is indicated in those patients whose outlook for a remission of their mental disease and ultimate discharge is reasonable. Also those patients who probably will not be able to leave the hospital but can adjust to minimum supervision and maximum privileges should be given serious consideration.

It is not believed that extensive surgical procedures are indicated in the chronic deteriorated psychotic patient whose psychiatric prognosis is hopeless and who must spend the remainder of his life under close supervision. However, minor surgical procedures, such as pneumothorax, pneumoperitoneum, and phrenico-exeresis have been employed in this group of patients without difficulty.

The active participation of the psychiatrist in the selection of cases for collapse therapy is an absolute necessity. This is particularly true for those patients who are candidates for major thoracic surgery.

In the actively psychotic patient with active pulmonary tuberculosis the decision must be made as to which illness to treat with the greater emphasis. The disturbed, hyperactive patient cannot be made to observe bed-rest to any degree. He also is unable to cooperate adequately with specific forms of treatment, and is difficult to manage during the postoperative period, if surgery is undertaken. This was well illustrated by one patient in the present series who expired following a second stage thoracoplasty. A brief summary of this case follows.

C. E. H., age 52, was admitted to this hospital March 27, 1946. The history revealed that the patient had been a chronic alcoholic for most of his adult life and in 1944 he was admitted to a state hospital in California following a long period of drinking. A diagnosis of psychosis, alcoholism, alcoholic neuritis and Korsakoff's syndrome was made. The patient failed to show improvement and was transferred to this hospital. On admission he was completely disoriented and showed marked impairment of memory for both recent and remote events. He later developed delusional ideas and was continually hyperactive, requiring frequent sedation and mechanical restraints. No improvement in his mental condition was noted during his hospitalization and on June 12, 1947 his diagnosis was changed to psychosis, alcoholic deterioration. Roentgenographic examination of the chest on admission to the hospital showed pulmonary infiltration in the right apex and first interspace. No cavities were seen. Gastric lavages were positive for the presence of acid-fast bacilli. Pneumothorax on the right was instituted February 3, 1947. Roentgenogram of the chest following institution of pneumothorax revealed the presence of pleural adhesions and a closed intrapleural pneumonolysis was attempted on March 25, 1947, but was unsuccessful. Thoracoplasty on the right was recommended and the first stage was performed on May 28, 1947 and the first three ribs were resected. The anesthesia used was nitrous oxide and oxygen and sodium pentothal intravenously. The patient's postoperative course was uneventful except for the difficulties experienced as a result of his mental condition. He seemed completely unaware that he had had an operation. He insisted on being out of bed immediately and used his arm on the operated side without apparent pain or discomfort. He demanded and consumed large quantities of food. Immediately postoperatively considerable sedation was required in order to control the patient. On the 25th of June, 1947 second stage thoracoplasty was performed and the fourth, fifth, sixth, and seventh ribs were resected. The

patient's immediate postoperative condition was good. On the first postoperative day he developed considerable abdominal distension accompanied by respiratory difficulty. Improvement was noted after tap water enemas and injection of prostigmine and the use of an oxygen tent. On the second postoperative day his condition was much improved and the oxygen was discontinued. At 6:25 p.m. on the second postoperative day patient sat on the side of bed and drank some coffee. After a few minutes he suddenly fell back on the bed, gasped, and respirations were observed to have ceased. Although postmortem examination was not performed and the exact cause of death remains obscure, it is believed that the patient's hyperactivity and general psychiatric status interfered in the proper management of his care.

It is now believed that an attempt should be made to improve the psychiatric condition of such patients before attempting major surgery. Shock therapy or prefrontal leukotomy would appear to offer the greatest possibility for a remission in the patient's psychosis.

In the past, pulmonary tuberculosis, active or arrested, has been generally considered a contraindication to shock therapy. More recently, however, a more liberal attitude has been adopted by some people. Moore (13) states that when proper precautions are followed and where the need is sufficient, latent, arrested, or even active pulmonary tuberculosis does not constitute an absolute contraindication to shock therapy.

The opinion expressed by Wilcox (14) in regard to the contraindications to shock therapy in general is felt to be worthy of emphasis and is quoted here: "The contraindications to shock therapy are related to the complications which may occur but are modified by the skill of the physician in preventing the complications and by the relative importance attached to the complications. Obviously, the more severe the mental illness the greater the risks that will be tolerated in the hope of therapeutic gain, but as the therapy is applied to milder cases the possible complications must be weighed even more carefully."

Certainly in many psychotic patients with pulmonary tuberculosis, the psychosis is more of a detriment to recovery than would be the case with shock therapy.

Reports of cases with pulmonary tuberculosis that have been given shock therapy are meager. Bulley and Greene (15) report one patient with an acute recent psychosis and active tuberculosis who was given metrazol shock, with improvement and subsequent arrest of the tuberculosis.

At the present time two patients on the writers' service are receiving electric shock therapy with curare. One of these patients has arrested minimal tuberculosis, and the other has a moderately advanced lesion that is active but has been stable for six months. Both patients have been under treatment for too short a time for any conclusions to be drawn as to the effect of electric shock therapy on the pulmonary lesions.

In the hands of an experienced neurosurgeon prefrontal leukotomy does not constitute a serious threat to the patient even if active pulmonary tuberculosis is present. One patient with far advanced pulmonary tuberculosis, who had been psychotic for many years, had a prefrontal leukotomy, following which a two stage thoracoplasty was performed in conjunction with streptomycin

therapy. His progress thus far has been satisfactory, and his outlook for recovery definitely improved. It is believed that the surgical therapy of the tuberculosis would not have been possible without the preceding prefrontal leukotomy. A brief case summary follows:

P. L. H., age 57, was admitted to this hospital on December 13, 1945. The patient had shown evidence of severe mental illness for many years and had been diagnosed as schizophrenic reaction of the hebephrenic type. A roentgenographic examination of the chest revealed the presence of a minimal infiltration in the right lung. The left lung showed a chronic proliferative inflammatory process extending from the second to the seventh ribs anteriorly. A cavity appeared to be present in the left second interspace. No active therapy was undertaken at this time. Reexamination in February, 1947 again revealed the previously reported pulmonary infiltration. There appeared to have been very slight regression since December, 1945. Investigations were carried out to determine the etiology of the pulmonary lesion demonstrated on the roentgenogram. The erythrocyte sedimentation rate was found to be 32 mm. per hour. The patient was unable to cooperate in producing sputum specimens and gastric lavage could only be performed by restraining the patient. The few lavage specimens obtained at this time were reported as negative for tubercle bacilli. The patient's mental disease was characterized by many bizarre delusions in reference to the genito-urinary and gastro-intestinal tracts. He also had many delusions of a religious nature. These abnormal thought processes were so disturbing that the patient required very close supervision and sedation. He refused to allow the windows to be opened near his bed, objected to any light in the room, and demanded that the other patients leave the radio off at all times. He was so insistent in these demands that he was involved in frequent fights with other patients. In view of the severity and long-standing nature of the patient's mental illness and the presence of undoubtedly active pulmonary tuberculosis, the patient's case was reviewed by the medical and psychiatric Staff. On their recommendation, a prefrontal leukotomy was performed on June 18, 1947. Postoperatively a marked improvement in his attitude and behavior was noted. He was cheerful, became interested in the ward activities, and cooperated fairly well with bed-rest and diagnostic procedures. More gastric lavages were performed and were reported as positive for the presence of acid-fast bacilli, thus establishing the diagnosis of active pulmonary tuberculosis. The patient's mental improvement continued and was felt to be such that a trial visit could be anticipated if his pulmonary tuberculosis could be brought under control. With this in view, pneumothorax on the left was initiated on August 5, 1947. It was discontinued, however, on August 19, 1947, because of the presence of pleural adhesions that were unsuitable for pneumonolysis. The patient's case was then reviewed by the Assistant Branch Chief of Tuberculosis and a thoracoplasty on the left was recommended. Streptomycin was to be given in conjunction with the thoracic surgery. The patient was started on streptomycin December 15, 1947, 0.5 Gm. twice a day. The first stage thoracoplasty was performed on February 11, 1948 and the first three ribs were resected. The anesthesia consisted of intravenous sodium pentothal, nitrous oxide and oxygen and curare. The patient withstood the procedure well and the postoperative period was free of complications. The second stage thoracoplasty was performed on March 10, 1948 and the fourth, fifth, sixth, and seventh ribs were resected. Again the patient withstood the procedure well and the immediate postoperative period was free from complications. At the present time the patient has shown some exacerbation of his mental disease. His attitude is one of antagonism and quarrelsomeness. It is felt, however, that, as his general physical condition improves and his feeling of well-being returns, his mental condition will again show improvement. Roentgenographic examination of the chest shows an adequate collapse of the diseased portion of the left lung. The minimal infiltration noted in the right lung on admission has remained stable. It is believed that this case shows that one can salvage at least a small percentage of patients with a disturbing psychosis and pulmonary tuberculosis.

Although this patient's case is not closed at the present, it is believed that his outlook has been definitely improved.

The institution of psychiatric treatment of the sort discussed above is not always necessary before starting such collapse measures as pneumothorax, pneumoperitoneum or phrenic nerve operations. In none of the 33 patients in whom pneumothorax has been started has it been necessary to discontinue the treatment because of the psychiatric condition. Altshuler and Bailey (16) also report that, of 100 patients treated with pneumothorax, only one was unable to continue to receive the treatment because of uncooperativeness.

In the cases suitable for pneumothorax, it is advisable first to establish the pneumothorax if possible and then to proceed with specific therapy for the mental disease, such as shock therapy or prefrontal leukotomy. The period of time necessary before proceeding with shock or prefrontal leukotomy varies with the individual case. In the disturbed hyperactive patient, measures should be taken as soon as possible in order to improve the patient to the point that he can at least accept a modified bed-rest regimen. On the other hand, it would probably be justified to wait until the patient's pulmonary tuberculosis is partially controlled in those patients who are quiet and fairly cooperative (catatonic, depressed). It should be remembered that the per cent of remissions following all forms of shock therapy decreases rapidly as the duration of the psychosis increases.

The writers present the following outline of a program for the care of the psychotic patient with pulmonary tuberculosis. It is realized that the presentation of such a program carries the risk of over simplification of the problem since each case must be considered individually.

The patients are divided into three large groups: A, B and C.

Group A patients include those whose psychosis is of recent origin and is of a type which may be expected to improve with specific therapy. If evaluation of the patient's pulmonary lesion indicates that collapse therapy should be undertaken, two courses of action are considered. In the disturbed hyperactive and uncooperative patients in which suitable collapse therapy cannot be instituted because of the patient's mental condition, it is advisable to proceed with specific therapy directed to alleviate the mental illness (*i.e.*, shock therapy, prefrontal leukotomy, or antisyphilitic therapy). Collapse therapy is then instituted as soon as the patient's mental condition is such that he can cooperate to a sufficient degree.

In the less disturbed, more cooperative patients in whom collapse therapy is indicated, it can be started first and then followed at a later date by specific treatment of the mental disease.

Group B patients are those whose psychosis is of a chronic nature and of such a severity that they are unable to leave the hospital but can adjust to minimum supervision. Many of these patients may be considered mentally competent as far as their cooperation in any treatment program is concerned. Depending upon the individual patient, collapse therapy is carried out in the same way as with a similar group of mentally competent patients.

Group C patients include those who have a psychosis of a chronic nature and who are either chronically disturbed or show marked mental regression and deterioration. It is this group of patients in which the highest incidence of pulmonary tuberculosis is found. Although it may be possible to carry out collapse therapy in a few of these patients, it is the exception rather than the rule. In general, no specific treatment is indicated except segregation from the hospital population. It is, however, in this group that prefrontal leukotomy should always be considered.

Thoracoplasty

The first thoracoplasty was performed at this institution in November, 1946. Since that time the procedure has been completed in 11 patients. Detailed information concerning these patients is presented in table 1. The youngest patient was 25 and the oldest 58 and the average age for the group was 47 years. Seven of the 11 patients had a trial of pneumothorax prior to thoracoplasty. In all 11 patients, pneumothorax was discontinued because of the presence of pleural adhesions unsuitable for pneumonolysis. The patients selected for thoracoplasty, with few exceptions, were those whose psychiatric prognosis was fairly good or those who had been able to make a satisfactory hospital adjustment (*i.e.*, open ward).

The operations were all performed by the local Consultant in Thoracic Surgery. The anesthesia has been of two types, local and general. The patients operated under local (1 per cent novocaine) received luminal 0.3 Gm., morphine .010 or .015 Gm. and hyoscine .0006 Gm. as a basal anesthetic. Five patients were operated upon under local anesthesia but only patients that were of fair mental competence or had marked catatonic features were selected for this type of anesthesia. All of the patients who received only local anesthesia were managed with difficulty during the operations.

General anesthesia was used in the remaining 6 patients, and consisted of intravenous sodium pentothal, nitrous oxide and oxygen and curare, with endotracheal intubation. All uncooperative disturbed cases were done under general as described.

There were two postoperative deaths in the group. The clinical course of one, C.H., was summarized above. The other, J.E.E., died on the third postoperative day. Postmortem examination revealed thrombosis of the superior sagital sinus and its main tributaries, with multiple hemorrhagic infarcts of the brain.

Although sufficient time has not elapsed for final disposition of the 9 surviving patients, 3 are being considered for trial visit, 4 are to be transferred to a ward allowing them to lead a more normal life, one has been recommended for prefrontal leukotomy, and the psychiatric status of one patient is such that continued close supervision will be necessary.

Pneumothorax

As previously noted, some physicians have been extremely pessimistic in regard to the use of pneumothorax in psychotic patients. Thirty-three of the

TABLE 1

Thoracoplasty cases

NAME	AGE	EXTENT OF PULMONARY DISEASE	NEUROPSYCHIATRIC DIAGNOSIS AND STATUS	DATE THORACOPLASTY COMPLETED	ANAESTHESIA	COMPLICATIONS	DISPOSITION
C. E. H.	52	M. A.	Psychosis, alcoholic deterioration, chronically disturbed, hyperactive. Requiring restraint and sedation.	Second stage 6–25–47	General	Died on second postoperative day.	
P. L. H.	57	F. A.	Schizophrenic reaction, hebephrenic type. Chronically disturbed, hyperactive. Requiring sedation and occasionally restraint. Prefrontal leukotomy performed prior to thoracoplasty with good result.	Second stage 3–10–48	General	Feeding problem. Requiring prolonged parenteral medication	Trial visit contemplated at the end of postoperative period of observation.
W. A. S.	55	M. A.	Paresis, in partial remission. Well adjusted hospital patient.	Second stage 2–12–47	Basal and local	None	Discharge by transfer to Old Soldiers' Home.
O. D. M.	29	M. A.	Schizophrenic reaction, hebephrenic type. Seclusive, paranoid, occasionally hyperactive and a feeding problem.	Second stage 4–10–47	Basal and local	Continued to be a feeding problem during postoperative period. Tube feeding required.	Except for poor food intake, patient showed improvement in mental condition. Trial visit contemplated.
J. V. S.	25	MIN	Schizophrenic reaction, hebephrenic type. Seclusive, regressed, manneristic, occasionally hyperactive.	Second stage 12–17–47	General	None	No change in mental condition. Continued hospitalization necessary. To be transferred to another ward when postoperative observation complete.

F. R. R.	47	M. A.	Schizophrenic reaction, hebephrenic type. Seclusive but a well adjusted hospital patient.	Second stage 2-17-48	General	None	Trial visit contemplated at the end of postoperative period of observation.
J. E. E.	58	M. A.	Schizophrenic reaction, hebephrenic type. Seclusive and delusional.	First stage 7-23-47	General	Died on second postoperative day.	
A. C.	51	M. A.	Paresis, delusional, hypochondriacal.	Second stage 8-13-47	Basal and local	None	Increased privileges. Trial visit contemplated.
A. L. W.	53	M. A.	Schizophrenic reaction, paranoid type. Depressed, seclusive, delusional.	Second stage 4-11-47	Basal and local	None	Prefrontal leukotomy planned.
E. C. K.	52	M. A.	Schizophrenic reaction, hebephrenic type. Regressed, delusional, feeding problem.	Second stage 4- 7-48	General	Feeding problem. Spread of lesion after first stage. Patient given streptomycin before proceeding with second stage.	Mental condition requires continued hospitalization under close supervision.
R. S. I.	52	M. A.	Schizophrenic reaction, hebephrenic type. Paranoid and seclusive.	Second stage 1-29-47	Basal and local	None	Open ward privileges.

patients in the present series have been given a trial of pneumothorax without undue difficulty. No sedative therapy was considered to have been necessary in these patients. It is recognized, however, that in some patients sedative therapy would be indicated. It is believed that pneumothorax cannot be given to certain patients, such as the extreme paranoid or agitated manic patient, but these will be the exception rather than the general rule. Moreover, if collapse therapy is indicated in such a patient, the possibility of performing prefrontal leukotomy or electric shock treatment before the collapse therapy should be considered in order to give better cooperation.

Summarization of table 2 indicates the following. An attempt was made to establish thirty-seven pneumothoraces. Five pneumothoraces were successfully instituted without pneumonolysis and eleven were successfully instituted with the aid of intrapleural pneumonolysis. Two bilateral pneumothoraces were started and maintained. Twenty-one pneumothoraces were unsuccessful because of adhesions that prevented therapeutic collapse and were not amenable to intrapleural pneumonolysis. There have been no complications in the group which did not require pneumonolysis. Only one complication was encountered in the unsuccessful group. Six complications occurred in the group requiring intrapleural pneumonolysis: adhesive pleuritis, 1; pleural effusion, 3; spread of disease, 1; and traumatic pneumothorax with massive collapse and death, 1. Four of the patients in this group of 33 have died. The death of only one was related to pneumothorax therapy.

It is recognized that these results are not as favorable as some published series; nevertheless, the failure to achieve better results cannot be attributed to the patients' psychoses. The failure to achieve better results is principally a consequence of the long duration of disease prior to the attempts at collapse therapy.

The patients' psychoses have not constituted a serious problem in management even in regard to intrapleural pneumonolysis. This surgical procedure can be successfully performed, even in the agitated patient, by use of general anesthesia with sodium pentothal. Cooperation during the postoperative period can be obtained by means of sedation and special nursing care. The "special" nurse is a factor of great importance in the postoperative course of any psychotic patient. Constant observation, untiring patience, persuasive charm, and knowledge of the importance of the compliance with postoperative orders are requisite in every "special" nurse.

It is the opinion of some that pneumothorax is not a worthwhile procedure in older age groups. Attention should be drawn to the fact that it was possible to institute pneumothorax successfully in 12 patients 45 years of age or older. From this limited experience it is believed that collapse can be successfully instituted in older age groups far more often than is generally realized. It is impossible to predict success or failure accurately and the attempt at collapse therapy should be made if the patient is considered a suitable candidate for pneumothorax.

One final fact can be demonstrated by analysis of this group. Seventeen of

the 33 patients contracted pulmonary tuberculosis after entering this hospital. This demonstrates the striking need for better control of tuberculosis in mental hospitals.

Other Collapse Measures

Phrenico-exeresis and pneumoperitoneum have also been found applicable in psychotic patients. The former procedure has been performed in 5 patients. Phrenico-exeresis has been reserved for situations in which it is desired to give the diseased lung additional protection against reactivation or extension of disease. The results have been equivocal.

Pneumoperitoneum has been instituted in two patients without difficulty. The concept has been followed that this form of therapy is indicated in patients with bilateral disease whose lesions are predominantly exudative in character. Pneumoperitoneum may also be used in certain selected patients who would be candidates for major chest surgery except for their psychiatric status. The following case history is cited to demonstrate such a patient.

J. A.S., age 53, was admitted to this hospital September 4, 1938. A diagnosis of psychosis, syphilitic meningo-encephalitic type, was made on admission. The syphilologist believed that no further antisyphilitic therapy was indicated. The patient showed marked mental regression and deterioration. He was unable to carry on a normal conversation and his psychiatric prognosis seemed hopeless. First evidence of pulmonary tuberculosis was discovered on routine roentgenogram of the chest in November, 1945. At that time an area of pulmonary infiltration was observed in the right apex and first interspace. Since 1945 there has been a moderate increase in the extent of the lesion but no spread to the contralateral lung. Gastric lavages have been positive for the presence of acid-fast bacilli. Right pneumothorax was initiated in March, 1947. Collapse was inadequate because of multiple adhesions which did not appear to be amenable to pneumonolysis. Therefore, pneumothorax was discontinued. The patient's psychiatric condition was so bad and his physical condition was so poor that thoracoplasty was thought to be contraindicated. He was tube-fed from July, 1947 through October, 1947 and his general physical condition improved. Pneumoperitoneum was begun in September, 1947. Recent roentgenographic examination of the chest revealed definite decrease in the extent of the infiltration in the lung. Phrenico-exeresis on the right is being considered at the present time to increase the elevation of the right diaphragm.

SUMMARY

The purpose of this paper has been to show that problems in the management of psychotic patients with pulmonary tuberculosis are not insurmountable. The fact that an appreciable number of psychotics make at least a partial recovery and return to their families and home communities and the problem of control of the disease in the institution make a more active approach desirable.

Conclusions based on experience with over 100 psychotic patients with tuberculosis are reported. Routine roentgenographic survey of the patient population has been proved to be the best method of detection of cases. General management of these patients has been discussed in detail with special emphasis on the problems of rest, nutrition, and collapse therapy. All forms of collapse therapy can be successfully carried out in selected psychotic patients. In

TABLE 2

Pneumothorax cases

NAME	KNOWN DURATION DISEASE	EXTENT OF DISEASE	AGE	PNEUMOTHORAX SUCCESSFUL WITHOUT AID	PNEUMOTHORAX SUCCESSFUL WITH PNEUMONOLYSIS	PNEUMOTHORAX UNSUCCESSFUL (ADHESIONS)	COMPLICATIONS	NEUROPSYCHIATRIC DIAGNOSIS AND STATUS
	yrs.							
N. S. A.	4	Min. uni-lateral	56			Yes	None	Schizophrenic reaction, catatonic type. Depressed, seclusive.
A. S. C.	11	MA bilateral (left, obsolete fibrosis)	48	(a) right, yes			None	Schizophrenic reaction, paranoid type. Delusional, occasionally disturbed and hyperactive.
A. C.	2	MA uni-lateral	51			Yes	None	Psychosis, syphilis, meningoencephalitic type. Delusional, hypochondriacal.
A. D.	4	MA uni-lateral	46			Yes	None	Schizophrenic reaction, simple type. Hypochondriacal, occasionally disturbed.
A. G.	1½	MA bilateral	57		(a) right, yes	(a) left, yes	None None	Psychosis, epileptic deterioration. Paranoid, argumentative, occasionally combative.
J. G.	2	MA uni-lateral	28		Yes		Adhesive pleuritis. Pneumothorax discontinued.	Schizophrenic reaction, catatonic type. Seclusive, manneristic.
W. L. H.	5	Min uni-lateral	51			Yes	None	Schizophrenic reaction, hebephrenic type. Deteriorated, manneristic, occasionally hyperactive.

F. C. H.	4	MA bilateral	55		(a) right, yes		Pleural effusion. Pneumothorax continued; effusion controlled by thoracentesis.	Schizophrenic reaction, hebephrenic type. Deteriorated, manneristic, compulsive.
J. K.	29	MA uni-lateral	56			Yes	None	Schizophrenic reaction, catatonic type. Seclusive, mute.
E. J. K.	3	MA uni-lateral	57	Yes			None	Psychosis, syphilis, meningo-encephalitic type. Seclusive, delusional.
W. C. K.	10	MA bilateral	60	(a) right, yes (a) left, yes			None None	Schizophrenic reaction, hebephrenic type. Seclusive, delusional.
W. H. L.	2	FA bilateral	51		(a) left, yes		Pleural effusion with pocketing. Pneumothorax discontinued.	Schizophrenic reaction, hebephrenic type. Deteriorated, delusional, occasionally hyperactive.
O. D. M.	2	MA uni-lateral	28			Yes	None	Schizophrenic reaction, hebephrenic type. Seclusive, paranoid, occasionally hyperactive and a feeding problem.
R. H. M.	14	Min uni-lateral	48			Yes	None	Schizophrenic reaction, hebephrenic type. Seclusive, deteriorated.
L. R. M.	2	MA uni-lateral	57		Yes		None	Schizophrenic reaction, hebephrenic type. Delusional.
U. L. N.	9	MA uni-lateral	50			Yes	None	Psychosis, syphilis, meningoencephalitic type. Delusional.

TABLE 2—*Continued*

NAME	KNOWN DURATION DISEASE	EXTENT OF DISEASE	AGE	PNEUMOTHORAX SUCCESSFUL WITHOUT AID	PNEUMOTHORAX SUCCESSFUL WITH PNEUMONOLYSIS	PNEUMOTHORAX UNSUCCESSFUL (ADHESIONS)	COMPLICATIONS	NEUROPSYCHIATRIC DIAGNOSIS AND STATUS
	yrs.							
J. D. P.	2	MA bilateral (left, obsolete fibrosis)	53			(a) right, yes	None	Schizophrenic reaction, hebephrenic type. Effect not appropriate.
F. R. R.	17	MA bilateral (left, obsolete fibrosis)	47			(a) right, yes	None	Schizophrenic reaction, hebephrenic type. Seclusive but a well adjusted hospital patient.
J. H. S.	2	MA unilateral	52			Yes	None	Psychosis, syphilis, meningoencephalitic type. Deteriorated, regressed, feeding problem.
J. V. S.	2	Min unilateral	24			Yes	None	Schizophrenic reaction, mixed type. Seclusive, regressed, manneristic, occasionally hyperactive.
J. E. S.	5	MA bilateral (left, stable)	45	(a) right, yes			None	Schizophrenic reaction, hebephrenic type. Delusional, some deterioration.
S. S.	6	MA unilateral	59			Yes	None	Psychosis with mental deficiency.

J. S.	2	MA bilateral (right, stable)	53		(a) left, yes		None	Schizophrenic reaction, hebephrenic type. Delusional and excitable.
H. N. T.	1	MA bilateral	24		(a) left, yes (b) right, yes		Spread to right lung necessitating right pneumothorax.	Schizophrenic reaction, hebephrenic type. Deteriorated, hyperactive.
B. B.	9	Min uni-lateral	54		Yes		Pleural effusion.	Schizophrenic reaction, hebephrenic type. Deteriorated, hyperactive, regressed.
G. B.	13	FA bilateral (confined to upper lobes)	55		(a) right, yes	(a) left, yes	Patient expired following traumatic pneumothorax with massive collapse in presence of bilateral pneumothorax.	Schizophrenic reaction, hebephrenic type. Deteriorated, manneristic, regressed.
A. C.	15	MA uni-lateral	63			Yes	None	Psychosis, syphilis, meningo-encephalitic type. Seclusive, paranoid, resistive to therapy.
A. C. G.	2	MA uni-lateral	56			Yes	Tension pneumothorax with bronchopleural fistula and development of empyema.	Schizophrenic reaction, catatonic type. Seclusive, mute, occasionally agitated, a feeding problem.
P. L. H.	2	FA bilateral (right, healed min. lesion)	57			(a) left, yes	None	Schizophrenic reaction, hebephrenic type. Chronically disturbed and hyperactive, requiring sedation and occasional restraint.

TABLE 2—*Concluded,*

NAME	KNOWN DURATION DISEASE	EXTENT OF DISEASE	AGE	PNEUMOTHORAX SUCCESSFUL WITHOUT AID	PNEUMOTHORAX SUCCESSFUL WITH PNEUMONOLYSIS	PNEUMOTHORAX UNSUCCESSFUL (ADHESIONS)	COMPLICATIONS	NEUROPSYCHIATRIC DIAGNOSIS AND STATUS
C. H.	yrs. 1	MA uni-lateral	53			Yes	None	Psychosis, alcoholic deterioration, chronically disturbed and hyperactive, requiring restraint and sedation.
E. C. K.	11	MA uni-lateral	52			Yes	None	Schizophrenic reaction, hebephrenic type. Regressed, delusional, feeding problem.
L. L.	3	FA uni-lateral	54			Yes	None	Psychosis, syphilis, meningo-encephalitic type. Deteriorated, hyperactive.
F. G. M.	13	Min. uni-lateral	56		Yes		None	Psychosis, syphilis, meningo-encephalitic type. Some deterioration.

general, however, major thoracic surgery should be reserved for those patients with a favorable prognosis for remission of their psychoses or the ones who have made a satisfactory adjustment under maximum privileges and minimum supervision. Shock therapy and prefrontal leukotomy in an effort to improve the mental status are applicable in certain selected cases of psychotic tuberculous patients.

SUMARIO

La Asistencia de la Tuberculosis en los Psicóticos

Tiene por propósito este trabajo demostrar que no son insuperables los problemas planteados por los psicóticos que padecen de tuberculosis pulmonar. El hecho de que una proporción apreciable de los psicóticos se reponen por lo menos parcialmente y regresan a sus hogares y colectividades y el problema creado por la lucha contra la enfermedad en una institucion indican la conveniencia de un ataque más activo.

Las conclusiones presentadas básanse en las observaciones realizadas en más de 100 psicópatas con tuberculosis. Las encuestas radiográficas sistemáticas de los enfermos del establecimiento ha resultado ser el método mejor para el descubrimiento de casos. Discutese a fondo la asistencia general de esos sujetos, recalcando en particular los temas del descanso, la nutrición y la colapsoterapia. En los psicóticos seleccionados pueden ejecutarse con éxito todas las formas de la colapsoterapia; pero, en general, debe reservarse la cirugía torácica mayor para aquellos en los que el pronóstico para la remisión de la psicosis es favorable o que han logrado un ajuste satisfactoria con un máximo de libertad y un mínimo de vigilancia. En ciertos tuberculos psicóticos bien escogidos tienen su aplicación el choc terapéutico y la leucotomía prefrontal con mira a mejorar el estado mental.

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ON FULMINANT TUBERCULOUS SEPTICEMIA WITH LEUKOPENIA

WALTER PAGEL AND A. L. WOOLF[1]

INTRODUCTION

The term, fulminant tuberculous septicemia, is used to describe a rapid process of generalised tuberculosis which either fails to cause any grossly detectable lesions or produces multiple necrotic areas which do not resemble the classical structure of tuberculous foci, but are replete with tubercle bacilli. Only 11 such cases have been described in the literature and most of them showed evidence of primary abdominal infection associated with a characteristic leukopenia (1). The diagnosis of tuberculous septicemia in all of these cases is made from the fulminant clinical picture often resembling typhoid fever, in concert with the anatomical changes, rather than by the demonstration of tubercle bacilli in the circulating blood. The latter is still subject to technical difficulties and a positive result may be obtained in the terminal stages of pulmonary phthisis, without the presence of fulminating tuberculous septicemia.

In this hospital, 2 cases of fulminant tuberculous septicemia have been observed. One of these cases presented evidence of primary abdominal infection and belonged to the second category, *i.e.*, producing grossly detectable necrotic foci (1). The second case, to be reported below, is of special interest in that it originated in a straightforward primary pulmonary infection and belonged to the first category of cases without macroscopic lesions.

CASE REPORT

Clinical illness: The patient was a 56 year old man who had felt well until one month before admission, at which time he had fainted and fallen in the street. Improvement was slow and anorexia, disinterest, increasing deafness, and rash on face, arms and legs persisted. Five days before admission the patient was noted to have twitching of face.

Physical examination on admission to the hospital revealed a temperature of 101°F., a respiratory rate of 40 per minute and diffuse coarse rhonci over both chests. The total leukocyte count was 2,050 per cu. mm., of which 46 per cent were neutrophils. A chest roentgenogram revealed the shadows of a patchy consolidation and partial collapse of the right lower lobe. The clinical diagnosis was agranulocytosis and auricular flutter. The possibility that the patient also had psittacosis was considered.

The course of the illness was one of progressive physical and mental deterioration. The auricular flutter was converted to auricular fibrillation with a rapid ventricular rate. Crepitations were detected at the lung bases. The total leukocyte count fell to 250 per cu. mm. and the blood urea nitrogen was 73 mg. per 100 cc. The lungs became increasingly congested and slight anomalous ocular signs with inequality of pupils appeared just before death. The cell count of the cerebrospinal fluid increased from one to 156 per cu. mm. The patient died eight days after admission to the hospital.

Postmortem findings: Both lower lobes of the lungs were consolidated and hemorrhagic, with the process being more pronounced on the right. The other lobes were congested.

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A subpleural wedge-shaped caseous area 2 by 1 cm. was found in the anterior axillary aspect of the right lower lobe (figure 1). The lymph nodes at the bifurcation of the trachea were very enlarged (7 by 5 cm.) and formed a solid hard white mass (figure 2). The paratracheal nodes were also enlarged. The liver weighed 1,520 Gm. and was pale. The spleen was dark and soft and contained a partly hemorrhagic infarct, 2.5 cm. in diameter, near the upper pole. The infarct was demarcated by a narrow yellowish white

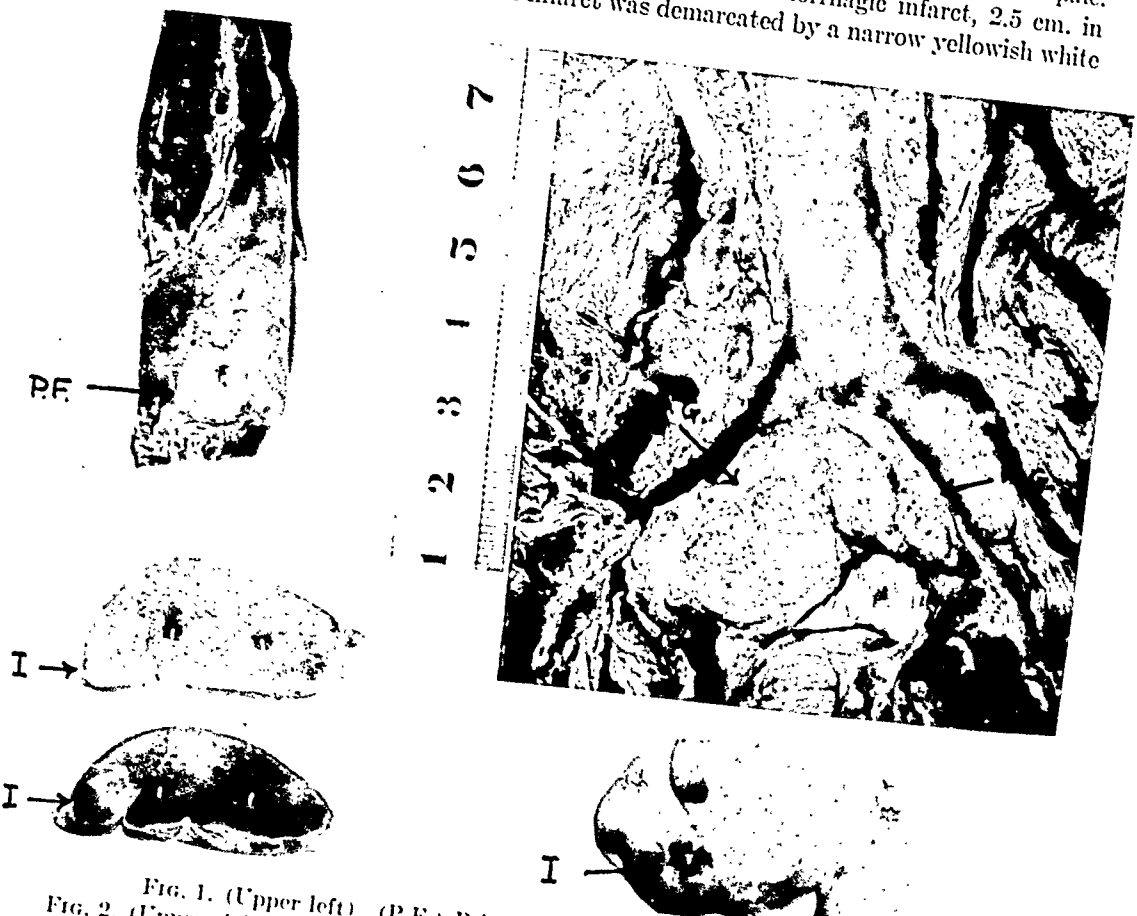


FIG. 1. (Upper left) (P.F.) Primary focus in the right lower lobe
FIG. 2. (Upper right) (P.G.) Confluent necrosis of the bifurcation lymph nodes, *i. e.* complementary primary lesion of the corresponding lymph nodes.
FIG. 3. (Lower left) Spleen with partly hemorrhagic infarction near the upper pole
FIG. 4. (Lower right) Kidney with small anemic infarct with hemorrhagic halo

zone (figure 3). There was a one-half centimeter in diameter anemic infarct with hemorrhagic halo (figure 4) towards the lower pole of the right kidney. There were raspberry-colored areas in the marrow of the femur while that of the sternum was a deep red.

Bacteriological investigations of postmortem material: Examination of a stained film of material from a bifurcation lymph node revealed numerous acid-fast bacilli. Cultures of material from bifurcation lymph nodes and splenic parenchyma yielded luxuriant growths of *M. tuberculosis*. Subcutaneous inoculation of a saline suspension of approximately 0.25 Gm. of macerated bifurcation lymph node into two guinea pigs caused

generalised tuberculosis which was discovered when they were killed forty-two days later. A rabbit inoculated with 0.001 mg. culture intravenously is still alive five months after infection.

It was concluded that a *human* strain of *M. tuberculosis* was responsible.

Careful postmortem roentgenography failed to reveal any trace of calcification in the lungs.

Histopathologic findings: Microscopic examination of the lungs revealed a diffuse hemorrhagic bronchopneumonia with much inflammatory edema but few cells. The latter were chiefly present in the alveolar phagocytes, some of which were filled with cocci. Polymorphonuclear leukocytes were scarce. The hemorrhages were chiefly visible in the periphery of the exudative lesions. Perivascular mononuclear cell infiltration and invasion of the intima of medium sized arteries were both present. Examination of other pneumonic areas revealed fibrinoid necrosis with central collections of cocci and peripheral hemorrhage. There were thickening and edema of arteriolar walls with mononuclear cell infiltration of the periphery of the adventitia. No acid-fast bacilli were demonstrable in appropriately stained sections of the hemorrhagic lesions.

Examination of the subpleural wedge-shaped caseous area revealed confluent caseous bronchopneumonia which was ill defined and showed very little, if any, evidence of encapsulation. The lesion was surrounded by a few lymphocytes distributed in the immediately adjacent atelectatic aveoli. The process appeared to be rapid and destructive as evidenced by fragments of bronchial cartilage and traces of vascular walls which could be seen in the peripheral part of the caseous area. Not more than two giant cells were found per section in the periphery of the focus and no classical tubercles were observed. The caseous area contained many acid-fast bacilli. Shadowy outlines of the elastic fibers of the alveolar septa were still visible but were entirely destroyed in those areas where there was beginning liquefaction. This change occurred in small patches and caused the usually pink caseous ground substance to assume a bluish tint presumably as a result of loss of the fibrinoid component.

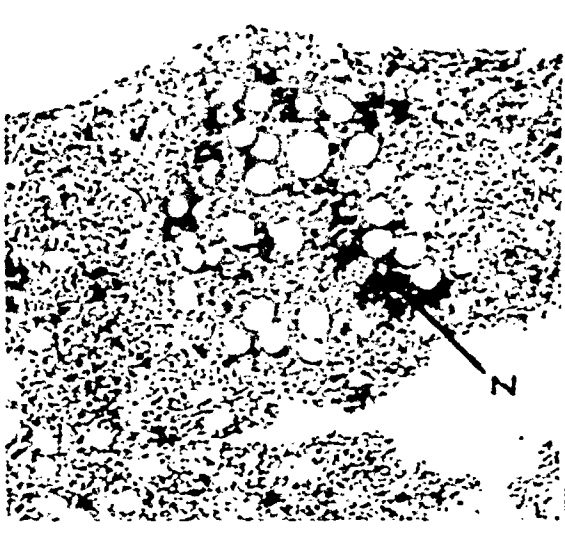
The bifurcation lymph node contained confluent ill defined patches of fibrinoid necrosis which consisted chiefly of round chromatin fragments.

The sternal marrow contained many cells, many acid-fast bacilli. No polymorphonuclear leukocytes were seen but many "plasma cells" were present. The marrow also contained fair numbers of immature red cells, an occasional eosinophile and other immature leukocytes and some megakaryocytes. In addition, there were a fair number of ill defined areas of fibrinoid necrosis containing many acid-fast bacilli (figure 5). These areas were surrounded by active bone marrow as described above and revealed no evidence that any attempt had been made to form zones of cellular demarcation (figure 6).

A very few small Malpighian corpuscles were visible in the spleen and the grossly hyperemic pulp dominated the picture. The pulp was moderately cellular and contained numerous reticulo-endothelial cells, especially in the sinuses. Some of these cells were full of hemosiderin (erythrophagia). In addition, scattered in the pulp, were small areas of fibrinoid necrosis with a moderate number of chromatin fragments, mostly round and rod shaped. These areas are ill defined, devoid of any attempt at demarcation and abound with acid-fast bacilli (figure 8). Finally, some vascular changes were noted, which consisted of a fibrinoid necrosis of the intima of the larger veins. These lesions contained numerous acid-fast bacilli (figure 7). The vascular changes were particularly prominent in and around the large, partly hemorrhagic, splenic infarct.

No acid-fast bacilli were discovered in the splenic tissue outside the small necrotic foci.

Multiple periportal as well as intralobular areas of fibinoid necrosis were present in the



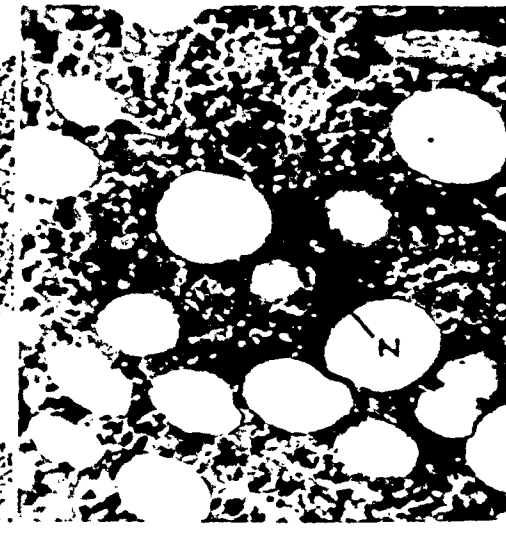


FIG. 5. (Left) Sternal marrow survey showing an area of fibrinoid necrosis (N) surrounded by collections of plasma cells mixed with scanty marrow cells. × 100.

FIG. 6. (Right) Close up view of area of fibrinoid necrosis (N) as shown in figure 5. Note plasma cells in the periphery of the necrotic area. × 300

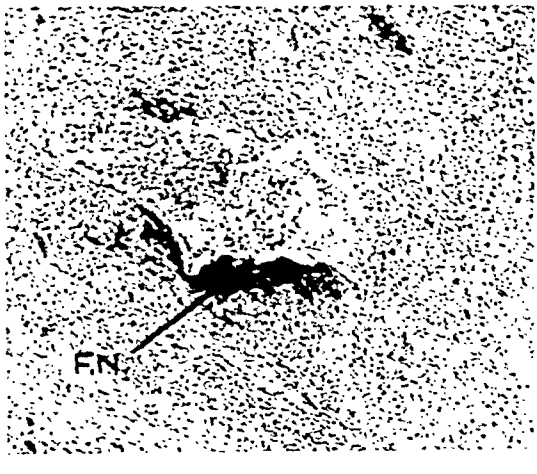


FIG. 7. (Left) Spleen: necrotising endophlebitis (F.N.) containing numerous tubercle bacilli (not shown). × 50.

FIG. 8. (Right) Spleen: from an area of necrosis. Ziehl Neelsen preparation. Mass of acid-fast bacilli. × 535.

liver. In the intralobular lesions proliferation of the Kupffer cells was prominent and many tubercle bacilli could be seen in the necrotic areas. There was an area of cortical necrosis in the kidney and the artery leading to the area was blocked by a thrombus. There were small areas of necrosis in the suprarenal cortex which contained moderate numbers of tubercle bacilli.

COMMENT

The wedge-shaped fresh caseous area in the lung, together with the wholesale necrosis of the bifurcation lymph nodes, constitute a primary complex, probably of short duration. Careful postmortem roentgenography of the lungs failed to show any trace of a preceding (now calcified) primary infection. It is therefore reasonable to conclude that the fresh lesion is of a primary nature in spite of the advanced age of the patient.

The process in this primary complex must have been rapid in view of the presence of "direct necrosis" rather than "caseation". The rapid nature of the necrosis is indicated by the predominance of *round* chromatin fragments, which originate from disintegrating lymphocytes and not from epithelioid cells, which leave long and bizarre nuclear remnants. The abundance of tubercle bacilli also points to a rapid process of tissue destruction.

Dissemination of tubercle bacilli, with resultant necroses, must have occurred from this primary lesion and corresponded to the five weeks of febrile illness. In spite of the microscopic areas of necrosis in the majority of organs, hardly any gross changes were visible. This is in contrast with the case previously reported from this hospital (1) and with the majority of cases recorded in the literature. Of the latter, only in the case reported by Scholz (2) were macroscopic changes not detectable.

The minimal gross changes in the present case included infarcts in the kidney and spleen. The former resulted from a nontuberculous arterial plug and the latter from tuberculous endophlebitis of one of the larger veins, the intima of which contained tubercle bacilli. A previous case reported by Pagel (3) was similar in that a large renal infarct and subserous petechiae were the only macroscopic findings.

Like the case previously reported from this service (1), the present case was characterised by a marked leukopenia attributable to the scarcity of leukocyte precursors in the marrow, and their replacement by plasma cells. Marked leukopenia (between 400 and 1,700 leukocytes per cu. mm.) has been described before, as a feature characteristic of tuberculous septicemia (2, 4, 5). In other cases, however, the total leukocyte count has been normal (6, 7, 8) or different blood changes were found, notably polycythemia (9), or acute myeloplastic leukemia with leukopenia (10). The case reported by Lenhartz (11) was one of a rapid miliary spread of only four weeks' clinical duration with the picture of acute myeloplastic leukemia. The similar observation by Crail (12), however, was chacterized by the long clinical course (about nine months) and splenectomy was performed.

The pulmonary portal of entry is consistent with a human tubercle bacillus infection, though it should be recalled that human bacilli have been recorded in

a number of cases of fulminant tuberculous septicemia in which the primary infection was abdominal. The latter cases seem more common than those following pulmonary infection. Only the cases described by Rennen (9), Balint (7), and Lenhartz (11), show evidence of a primary pulmonary infection.

SUMMARY

A case is reported of fulminant tuberculous septicemia of four weeks duration which was associated with a marked leukopenia. The portal of entry was a fresh pulmonary primary complex with direct necrosis, and tubercle bacilli of human type were isolated from the lesions. It is believed that this case belongs in the category of those rare cases in which virtually no gross changes are seen.

SUMARIO

Septicemia Tuberculosa Fulminante con Leucopenia

El caso comunicado es de septicemia tuberculosa fulminante de cuatro semanas de duración, asociada a hiperleucopenia. La vía de entrada fué un complejo pulmonar primario reciente con necrosis directa, aislándose de las lesiones bacilos tuberculosos de tipo humano.

Este caso corresponde a esa rara forma de infección tuberculosa en que el examen macroscópico apenas revela patología.

ACKNOWLEDGMENT

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AGRANULOCYTOSIS DURING THE STREPTOMYCIN TREATMENT OF MILIARY TUBERCULOSIS

Report of a Case

DAVID D. FELD[1]

INTRODUCTION

Toxic reactions to streptomycin have been recorded by numerous observers, but very little reference has been made to any deleterious effects upon the hematopoietic system. In a recent report (1) on streptomycin toxicity by Farrington and his associates, it is stated under miscellaneous observations: "Leukopenia without granulocytopenia appeared in two subjects in conjunction with the appearance of a drug sensitivity reaction. In another subject, leukopenia (2,000 to 3,000 cells per cu. mm.) developed during the second month of therapy. A relative granulocytopenia was also present for approximately sixty days but eventually disappeared." They were doubtful that this reaction was on a toxic basis because the patient was a radiologist who apparently always had a low total leucocyte count and he was also suffering from acute hematogenous tuberculosis.

In a report (2) to the Council on Pharmacy and Chemistry on the effects of streptomycin on tuberculosis in man over 900 treated cases collected from the Veterans Administration, the Army, and the Navy are considered and eight instances (.98 per cent) of blood dyscrasia appeared during treatment. Five of these consisted of a relatively mild leukopenia with neutropenia. There were three instances, however, of agranulocytosis which was definitely due to streptomycin. Although one of these 3 patients had generalized miliary tuberculosis, cessation of streptomycin was followed by a return to normal bone marrow function despite progression of the disease.

The instances of leukopenia and granulocytopenia mentioned in these reports have not been presented in detail. Accordingly, it was thought advisable to report the following case in which leukopenia and agranulocytosis occurred, apparently as a complication of streptomycin therapy.

CASE REPORT

M. W., a 42 year old white male steelworker, was admitted to Muirdale Sanatorium on the evening of February 27, 1947.

His past history revealed that he had had rheumatic fever as a child. He was told that he had rheumatic heart disease, and he stated he had a "heart murmur as long as I can remember." For the six years prior to admission he had imbibed alcohol freely and on December 10, 1946, he had been admitted to the Milwaukee County General Hospital for acute alcoholism. A routine chest roentgenogram at that time was reported as being normal. He was discharged thirteen days later with a diagnosis of alcoholic psychosis. He returned to work and immediately began to notice dyspnea, loss of

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appetite, weight and strength, chills and night sweats. He continued to work however, until February 18, 1947, at which time he developed a very severe epistaxis. He was admitted to a private hospital where the left nasopharynx was packed to control the bleeding. The pack was removed a number of times but had to be replaced because of persistent bleeding. A chest roentgenogram, obtained on February 27, 1947 because of persistently high temperature, revealed the characteristic findings of miliary tuberculosis and he was transferred to Muirdale Sanatorium.

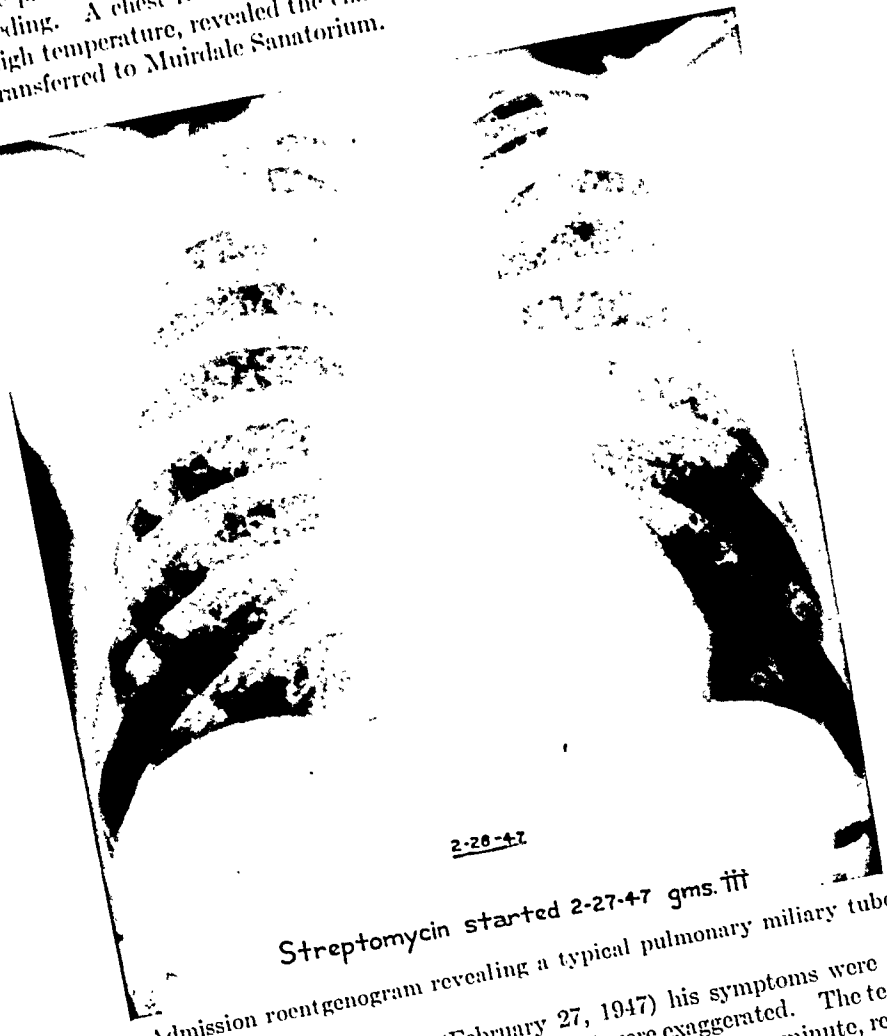

FIG. 1. Admission roentgenogram revealing a typical pulmonary miliary tuberculosis.

On admission to the sanatorium (February 27, 1947) his symptoms were similar to those that he had had at the onset of the disease, but were exaggerated. The temperature was 102.6°F. and the pulse and respiratory rates were 110 and 24 per minute, respectively. The physical examination revealed a fairly well developed and well nourished adult male (143 pounds) who appeared acutely ill. There was a nasal pack protruding from the left nostril which was dripping bright blood. The pack was removed three days after admission and no further bleeding occurred.

The significant findings upon examination of the chest were moist rales over the upper one-third anteriorly and posteriorly.

TABLE 1

DATE	HEMOGLOBIN PER CENT	COLOR INDEX	RBC MILLIONS/CU.MM.	WBC PER CU.MM.	DIFFERENTIAL WBC (EXPRESSED IN PER CENT)							PLATELETS THOUSANDS/CU.MM.	N.P.N. γ PER CENT	REMARKS
					NEUTROPHILS	SEGMENTED	STAB	LYMPHOCYTES	MONOCYTES	EOSINOPHILS	BASOPHILS			
Streptomycin started 2-27-47														
3- 1-47	71	.9	3.93	10,000	71	67	4	20	8	1				500 cc. citrated blood, March 3, 6, 10, and 24, 1947
3- 6-47	65	.93	3.56	7,050	77	69	8	16	4	1	2		29.5	
3-17-47	94	.98	4.88	7,850	57	55	2	22	15	6				
3-22-47	81	.92	4.46	3,100	26	23	3	53	19	0	2			
3-24-47				2,700	0	0	0	65	26	9				Neutrophils absent
3-25-47	Streptomycin stopped			2,250	1	1		50	39	9	1			Neutrophil degenerate
3-26-47				2,325	1		1	55	33	10	1			
3-27-47				2,375	1		1	62		6	2		51.2	
3-28-47				2,150	2		2	64	28	3	3			
3-29-47	87.7	.9	4.80	2,950	3		3	71	26			230		
3-31-47				2,825	19	11	8	52	29					
4- 1-47				2,800	20	7	13	49	28	3				
4- 2-47				3,950	38	19	21	37	22		1			
4- 3-47				5,300	57	34	23	28	15					
4- 4-47				7,350	58	38	20	29	13					
4- 5-47				10,450	67	48	19	20	13				40	
4- 7-47	87	.87	5.06	12,550	79	66	13	13	8					
4- 9-47				13,950	77	64	13	14	8		1			
4-11-47				17,150	77	61	16	12	11					
4-12-47	Streptomycin started			15,950	77	66	11	14	9				48	
4-14-47				12,250	74	59	15	15	9	2				
4-17-47	87	.85	5.19	12,400	73	65	8	13	8	6			38.4	
4-30-47				12,150	73	63	10	16	7	4			39.5	
5- 5-47	94	1	4.68	10,900	64	58	6	28	2	4	2			
5-12-47				9,450	66	60	6	22	10	2			44	
5-20-47				10,300	68	60	8	21	6	5			45.4	
5-28-47				14,900	81	73	8	13	3	3				maxillary antrum infection
5-31-47				10,800	76	73	3	16	12					
6- 7-47				10,500	59	52	2	16	15				51.9	
7- 2-47				9,350	55	51	4	27	12	6				
7-30-47				9,250	48	44	4	35	15	3			50	
8-21-47	94	.85	5.66	10,650	56	49	7	26	15	3			26.6	
9- 2-47				9,900	61	57	4	30						
9-25-47				10,200	57	52	5	36	7				31.9	
10-27-47				9,100	54	45	9	37	6	3				
11-27-47	100	1	6.24	9,250	61	50	11	33		2				
12-26-47	100	1	5.99	10,200	53	50	3	36	7	3	1		39.4	
1-29-48	107	.97	5.51	8,550	56	51	5	38	5	1				
3- 8-48	113	.97	5.86	9,000	56	54	2	38	4	2		308	29	
4- 2-47	Bone marrow biopsy—tubercle formation. Streptomycin blood levels ranged from 2.5 to 5 γ per 100 cc. Total days treated—250. Total drug given—659.6 Gm.													

There was a presystolic rumble and a harsh mitral systolic murmur over the apex and left sternal margin of the heart. The blood pressure was 110/60 in mm. of mercury.

The liver was not tender and extended 5 cm. below the costal margin. The spleen was not palpable.

The chest roentgenogram (figure 1) revealed the characteristic densities of miliary tuberculosis.

Examination of a concentrated specimen of sputum revealed acid-fast bacilli on microscopic examination. A Vollmer patch test for tuberculin sensitivity was positive.

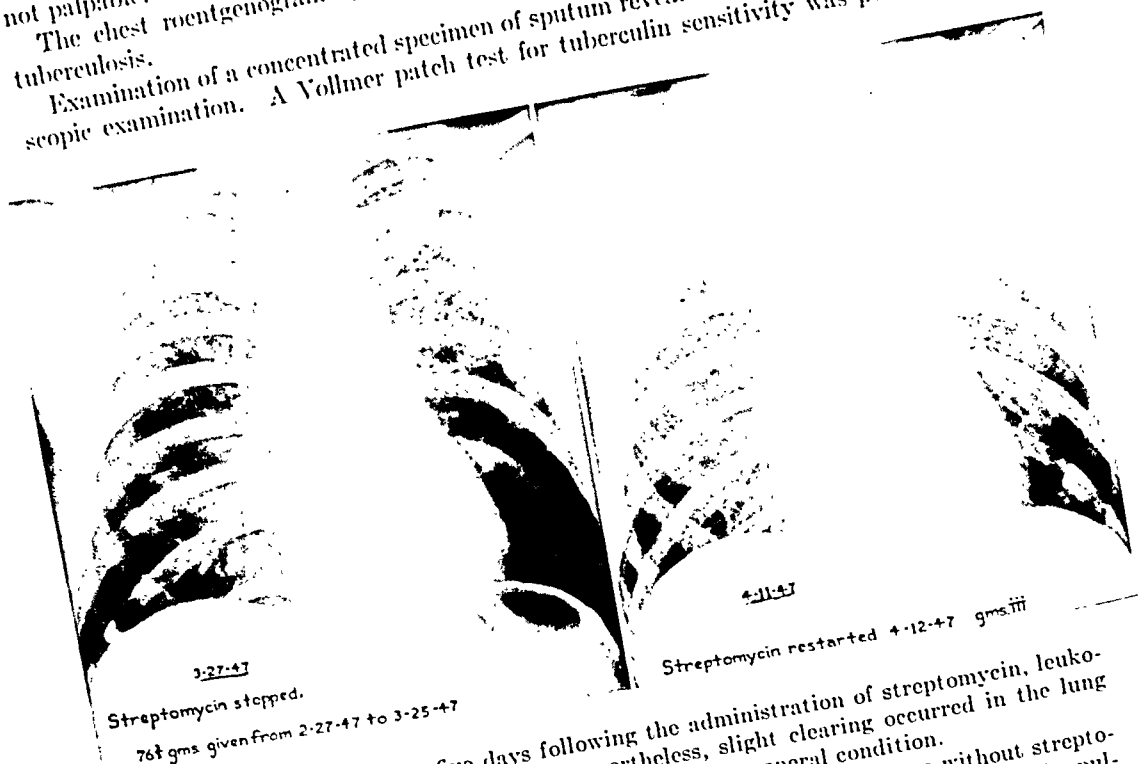

FIG. 2. (Left) Twenty-five days following the administration of streptomycin, leukopenia and agranulocytosis developed; nevertheless, slight clearing occurred in the lung fields and there was some improvement in the patient's general condition.

FIG. 3. (Right) Forty-three days after admission and sixteen days without streptomycin. Clinical condition is worse and there has been considerable increase in the pulmonary infiltration. However, the blood picture has definitely improved.

Therapy: Streptomycin therapy was started immediately and 3 Gm. were given every twenty-four hours, in divided doses at four hour intervals. The treatment was interrupted on the twenty-fifth day of March, 1947 because of leukopenia and agranulocytosis (table 1, figure 2). The total amount of drug which had been given was 76.6 Gm. Streptomycin from three different manufacturers was used during this period. At the time of the agranulocytosis the patient exhibited none of the other characteristic evidences of drug "hypersensitivity". Moreover, he denied ever having had episodes suggestive of allergic phenomena.

After a sixteen day interval, the daily dose of 3.0 Gm. of streptomycin was restarted April 12, 1947 and was continued until October 28, 1947, when it was reduced to 1.0 Gm. Streptomycin therapy was discontinued on November 23, 1947. The total number of days the patient was treated with streptomycin was 250 and the total amount of drug given was 659.6 Gm.

Other treatment consisted of: three blood transfusions of 500 cc. each of citrated blood given on March 6, 10, and 24, 1947; ferrous sulphate; and vitamin preparations. Two cc. of liver extract was given twice each week until April 21, 1947. The only other drugs given were Seconal, 0.09 Gm. (Sodium-propyl-methyl-carbinyl-allyl-barbiturate) from February 27, 1947 to March 30, 1947, a total of 1.5 Gm., and phenobarbital, 0.09 Gm. Since March 1947 there has been no other medication with the exception of an occasional dose of milk of magnesia.

Clinical course: The patient was very toxic and up to the time that streptomycin therapy was interrupted on the twenty-fifth day there had been no change in the temperature or pulse. There had been slight clinical improvement, however, and his appearance was better, his appetite had improved, and there was slight roentgenological improvement (figure 2).

During the sixteen day interval that the drug was withheld, the clinical condition of the patient grew worse, cough and expectoration increased and appetite decreased. A roentgenogram, taken at this time, revealed a definite increase in the pulmonary infiltration (figure 3).

In view of the patient's critical condition and the usually malignant nature of this type of tuberculosis, it was decided to reinstitute streptomycin therapy, using the previous dosage. Moreover, by the end of the sixteen day interval without chemotherapy, a high percentage of granulocytes were present in the peripheral blood (table 1). Streptomycin was reinstituted on April 12, 1947 and was followed by a gradual improvement in the patient's condition. On May 28, 1947 he developed an infection in the right antrum, associated with marked facial swelling and tenderness over this area. Penicillin therapy was started in addition to the streptomycin and within forty-eight hours the swelling and pain had completely disappeared. It should be noted that the added infection caused an appreciable elevation in the total leucocyte count with very little change in the percentage of granulocytes.

Improvement of the status of the tuberculosis continued, there was a weight gain of 30 pounds and during the six months after the cessation of streptomycin he had had no constitutional or local symptoms.[2]

The drug was given for a period, perhaps well beyond its benefits, but at that time experience in the streptomycin treatment of miliary tuberculosis was limited and the possibility of late relapse was well recognized.

In vitro studies of the streptomycin resistance of the patient's organisms could not be carried out as it was impossible to culture the tubercle bacilli. Obviously the infection was still very sensitive to the drug, however, even after twenty-five days of streptomycin treatment, as shown by the observed improvement when the drug was readministered.

Slight renal irritation, as indicated by occasional cylinduria, albuminuria and minimal azotemia were present, but disappeared completely after the cessation of chemotherapy. Very slight vestibular dysfunction was noticed for about the first three months of therapy, but numerous audiometric examinations revealed no abnormal findings.

COMMENT

Although miliary tuberculosis is recognized as being one of the etiological factors in leukopenia (3), the writer has never seen a case when leukopenia has been this marked or has been accompanied by neutropenia of the extreme degree noted in this patient.

[2] There has been no recurrence of the disease; patient remains in excellent condition.

In table 1 it should be noted that on March 24, 1947 and in four successive leucocyte counts, the neutrophils ranged from absent to one, one, two, and three cells, respectively. Eosinophils were always present, so the term agranulocytosis is in its strictest sense not correct. Nevertheless, the term has been used rather loosely (3) and can be applicable to this type of reaction.

Seconal (sodium propyl-methyl-carbinyl-allyl-barbiturate) and phenobarbital were given during the early phase of the disease. These drugs have never

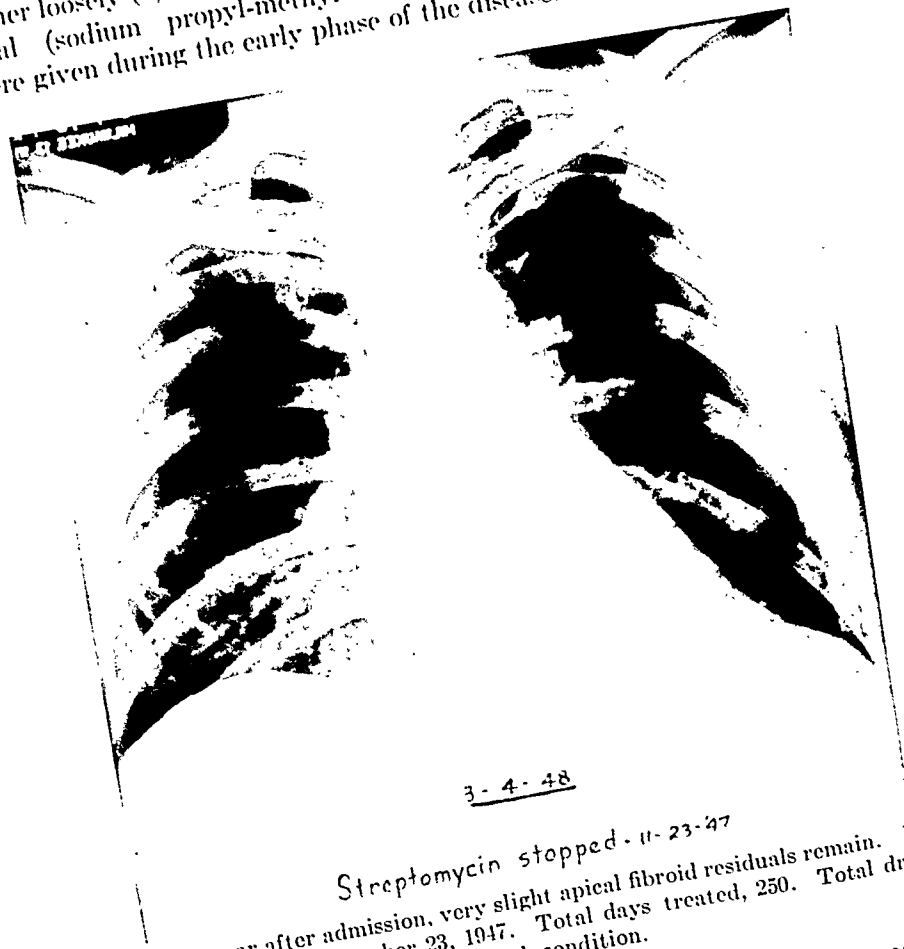

FIG. 4. One year after admission, very slight apical fibroid residuals remain. No streptomycin therapy since November 23, 1947. Total days treated, 250. Total drug given, 659.6 Gm. Patient is in excellent clinical condition.

been implicated as a cause of leukopenia or agranulocytosis. Damachek (3) states: "The barbiturates have been suspected many times, but rarely has there been clear-cut proof of their relationship to the disease agranulocytosis."

During the phase of agranulocytosis, there was no decided clinical change in the patient's condition but as evil consequences were feared, administration of the streptomycin was stopped. Perhaps the patient became densensitized to the drug during the interim, although when the drug was re-established, a slight drop in the total leucocyte count from the preceding levels was noted. This might have indicated incomplete desensitization. Sherman (4) in discussing

drug allergy in relation to agranulocytosis states that in some patients in whom agranulocytosis or severe leukopenia has been attributed to sulfonamides (4a) or thiouracil (4b), the same drug has been subsequently tolerated without any reaction.

During the past five months, frequent total leucocyte counts (not all recorded) have been normal. The total leucocyte count has ranged from 8,500 to 10,000 cells per cu. mm. with normal percentages of the various cell types.

Studies of the bone marrow, unfortunately, were not satisfactory, but examination of a specimen obtained by bone marrow biopsy on April 2, 1947 revealed the presence of tubercle formation and hence confirmed the diagnosis of generalized miliary tuberculosis.

Although the initial sputum examination was positive for acid-fast bacilli on direct examination, the organisms failed to grow on culture. Numerous cultures of sputum and fasting gastric contents obtained subsequently have all been negative. The virtually normal roentgenographic findings one year after admission may be seen in figure 4. At the time of writing the patient is asymptomatic and fully ambulatory.

SUMMARY

A case of miliary tuberculosis in which leukopenia and agranulocytosis appeared during streptomycin therapy is reported. The total amount of drug given up to the time of the agranulocytosis was 76.6 Gm. within a period of twenty-five days. At the height of the reaction no neutrophils could be detected on one examination of the blood and in five successive examinations the total neutrophil percentages were: one, one, two, and three in that order. When streptomycin therapy was interrupted, the patient became worse, as evidenced by an increase in symptoms and pulmonary infiltration, but there was a return of circulating neutrophils. Streptomycin therapy was re-established after an interval of sixteen days in the same daily dose (3.0 Gm.) as was used before the agranulocytosis. No further toxicity was noted and there was gradual clinical improvement. The patient has been completely asymptomatic for six months following the cessation of therapy.

It is believed that the marked neutropenia represented a toxic reaction to streptomycin (presumably allergic in nature) and was not merely a consequence of the generalized hematogenous tuberculosis.

SUMARIO

Caso de Granulia con Leucopenia y Agranulocitosis

1. El caso comunicado es de un enfermo con granulia que manifestó leucopenia y agranulocitosis durante una reacción de hipersensibilidad a la estreptomicina.

2. La dosis total de estreptomicina para la fecha en que apareció la agranulocitosis era de 76.6 Gm., administrados durante un período de 25 días.

3. Al interrumpirse la estreptomicinoterapia, la agranulocitosis desapareció. pero la tuberculosis avanzó, según demostraron el aumento de los síntomas y la extensión de las infiltraciones pulmonares.

4. La estreptomicinoterapia fué reanudada al cabo de 16 días. No se observó más toxicidad y mejoró gradualmente el estado de la infección tuberculosa.

5. Una biopsia de la médula ósea reveló formación de tubérculos, indicando diseminación miliar generalizada de la infección.

6. El enfermo recibió un total de 659.6 Gm. de estreptomicina, administrados durante un período de 250 días.

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A DIRECT METHOD OF STANDARDIZING DILUTE SUSPENSIONS OF TUBERCLE BACILLI BY USE OF A CAPILLARY MICROPIPETTE[1]

WILLIAM D. CRANDALL

INTRODUCTION

The precision of methods used in standardizing bacterial suspensions has been limited by the difficulty of resolving two chief sources of error: (a) the irregular dispersion of bacteria in fluids or in direct smear preparations; (b) the variability involved in computing densities of the order of fifty thousand to one billion organisms per cc. on the basis of small numbers of organisms visualized in small fractions of a limited number of small aliquots of the suspension, *i.e.*, the "Microscope Factor" of Frost (1).

The following investigation was undertaken in an effort to point out the limitations of the present methods and to devise a method permitting more precise standardization of dilute bacterial suspensions (those containing less than fifty million cells per cc.) by reducing the above mentioned sources of error.

Bacterial suspensions are widely used as vaccines, in serologic reactions, and in the investigation of the properties of bacteria as regards growth, electrical potentials, et cetera. Although in practice concentrations of bacteria are measured indirectly by reference to dry or moist weight (2), volume of centrifuged organisms (3), opacity (4 to 8), or density (9, 10), these methods in turn must be standardized by direct methods.

The chief direct methods are: colony counts of dilution plating, chamber counts, and counts of bacteria in stained smear preparations. Colony counts refer to viable cells. The minimal coefficient of variation of the dilution plating method has been postulated to be ±6.6 per cent (11). The main sources of error are well known (1, 12, 13). The wide range of variation in colony counts by plating methods in the case of the tubercle bacillus has been demonstrated by many investigators (6, 14 to 18).

Chamber counts to determine the concentration of tubercle bacilli were introduced in 1891 (19). Notable improvements in this method have been the use of a counting chamber with a well depth of 0.02 mm. by Mallory and Wright (20), the use of bacterial stains (20), the application of darkfield illumination (21), and finally the use of a chamber with a well depth of 0.01 mm. (22). Although Blumenberg (23) claimed an error of 1.2 per cent of the average count with a suspension of approximately 232 million organisms per cc., the error of this method has been considered to be between 5 and 10 per cent of the average count with suspensions of more than 50 million cells per cc. (7, 12, 20, 22), (table 1).

With suspensions of less than 50 million cells per cc. the error has been found to be about 50 per cent of the average count (12). This large degree of error is understandable in view of the fact that one optical field with the 0.02 mm. depth

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counting chamber represents one twenty-millionth of a cubic centimeter and theoretically would contain only 2.5 organisms if a suspension of 50 million cells per cc. were being examined. The law of small numbers (33) would require the counting of at least 50 such fields and even then the factor for computing the density of the suspension would be 400,000. Hence, the counting chamber is not applicable to the standardizing of dilute bacterial suspensions.

The limitations of standardizing suspensions of bacteria by counting stained organisms in smear preparations depend upon several well recognized factors (24). The errors inherent in sampling have led to the use of the 0.01 cc. delivery pipette (0.0101 ± 0.00008 cc.) in preference to the standard loop (0.0111 ± 0.0021 cc.) (24), or the dropping pipette (26 to 29) for accurate delivery of small

TABLE 1

Variations in counts by the counting chamber methods

	INVESTIGATOR		
	Wilson (12)	Blumenberg (23)	Steiner (22)
Depth of chamber	0.02 mm.	0.02 mm.	0.01 mm.
Optical field area	0.0025 mm.^{2}	0.000625 mm.^{2}	0.0025 mm.^{2}
Fraction of cc. in optical field area	$\frac{1}{20,000,000}$	$\frac{1}{80,000,000}$	$\frac{1}{40,000,000}$
Density of the suspension used	14,730 million cells per cm.^{3}	232 million cells per cm.^{3}	1,200 million cells per cm.^{3}
Average number of bacteria per field	700	2.67 to 2.9	28
Coefficient of variation of field counts	—	—	12 per cent (on basis of ten fields)
Coefficient of variation of counts and Number of fields on which count was based	3 to 5 per cent (120 fields)	1.2 to 2.3 per cent (50 fields)	6.4 per cent (10 fields)

volumes. Likewise, a dispersal area of 1 sq. cm. has been accepted in preference to larger areas as the error in sampling diminishes as the sample approaches 100 per cent of the whole. For example, one hundred samples of one thousand items are more truly representative than are ten samples. On the basis of time consumption, the upper limit of the area of a smear which it is practicable to count microscopically is about 0.06 sq. cm. (300 optical fields of 0.0002 sq. cm. each), requiring fifteen to twenty minutes. And as this area represents 6 per cent of the 1 sq. cm. smear but only 1.2 per cent of the 5 sq. cm. smear, it is more truly representative of the former smear than of the latter.

The error due to the variability of dispersion of bacteria in the dried smear (14, 25, 30) has recently been emphasized by Hanks and James (31). This variability, together with the requirements of large numbers of samples neces-

sary to attain reliable values when dealing with small numbers (32, 33), has no doubt prevented the stained smear counting method from attaining an acceptable degree of accuracy for anything except the roughest sort of an estimation. For example, if 0.1 cc. of a suspension of 33,333 tubercle bacilli per cc. was spread over a 5 sq. cm. area and an optical system with a field area of 0.015 mm.^{2} was employed so that 50 such fields represented 1/666 of the original 0.1 cc., then twenty counts of 50 fields each would yield total counts of from one to ten bacilli and would give a computed value of from 6,666 to 66,666 bacilli per cc. (32).

As examples of the degree of precision of the stained smear method, the following observations are of interest. Breed (24), using the 0.01 cc. volume spread over an area of 1 sq. cm. and counting 100 fields of 0.0003 sq. cm. each (1/3,000 of the sample), found a coefficient of variation of 59 per cent on counting 14 samples and of 49 per cent on counting 15 samples of a suspension containing 5,500 cells per cc. Duplicate counts on 3 suspensions containing 40 to 123 million cells per cc. showed coefficients of variation from 18 to 21 per cent.

Breed and Stocking (35), using similar preparations and counting the organisms in 200 fields of 0.00015 sq. cm. each (1/3,000 of the sample), found coefficients of variation of 10.5 to 17.3 per cent on counting 72 samples of each of 3 suspensions which contained from one to four million cells per cc. Duplicate counts on 35 suspensions with an average density of 250,000 cells per cc. showed a coefficient of variation of 1.2 per cent.

Nuttall (26) used a dropping burette to deliver drops of 0.006 to 0.01 cc. size onto a glass slide. He measured the number of optical fields across the stained drop preparation and computed the total number of fields. A count of 500 fields was then used to compute the density of the suspensions. With suspensions computed to contain 13 million to 3,400 million tubercle bacilli per cc. he found the difference of the count between two separate drops to be 0.7 to 10 per cent of the mean count.

It is to be noted that the Breed smear technique uses a microscopic factor of 3,000 which is decidedly less than the factor of 400,000 employed in the chamber counting method. Nuttall's method, by utilizing a smaller area of dispersal, together with counting of a much greater percentage of the sample, still further reduced the variability.

A means of circumventing the sampling errors involved in counting a limited number of fields in stained smears with irregularly distributed organisms was demonstrated by Ziesche (36) in 1907, who counted all of the stained organisms contained in the entire smear of droplets collected by exposing a glass plate in the field of a patient's cough.

Theoretically the advantages of the Breed smear and of Nuttall's technique could be combined by making a smear with 0.0003 cc. of a bacterial suspension. Such a smear would permit counting of all the organisms and thus would avoid the errors of irregular distribution and of microscopical sampling and would have a microscopic factor of 3,333. A pipette to deliver 7 cu. mm. with an error of less than 0.3 per cent was described by Linderstrom-Lang and Holter in 1931 (37) and a modification of this pipette permitting manipulation of volumes of 0.5 cu. mm. was presented by Claff (38) in 1947.

EXPERIMENTAL

Methods and Materials

With the advice and aid of W. L. Doyle and L. S. Sonkin, micropipettes were fashioned by pulling a very fine tip, without local constrictions, on short segments of capillary tube with a diameter of about 2.5 mm. and a uniform bore of about 0.24 mm. diameter. A strip of graph paper lined in millimeters was fastened on the back of each pipette. A short segment of mercury measured at various positions along the pipette demonstrated uniformity of the bore above the tapering tip. Comparison of the weight of 10 mercury specimens with the length of the bore which they had occupied determined that a 10 mm. length of the bore was equivalent to 0.475 cm. with a standard deviation of the mean of ± 0.002 mm.^{3}. Eleven samples of twenty normal sulphuric acid corresponding to 5 mm. lengths of the capillary bore were delivered into small dishes and titrated with one-tenth normal sodium hydroxide by S. Black, using an unpublished microtitration method. These titrations showed 5 mm. length of bore to be equivalent to 0.2297 cm. with a standard deviation of the mean of ± 0.0036 mm.^{3} delivered.

The apparatus for using the capillary micropipette is shown in figure 1. The micropipette (A) is held vertically by means of a one-hole rubber stopper and clamp (B) connected to a ring stand. A short section of stiff rubber tubing (C) connects the upper end of the pipette to one branch of a glass tube with two right angle branches (D) which also is supported by a clamp (E) attached to the ring stand. Compressed air from the laboratory line is passed through filter and wash bottles to remove dust and moisture and enters the glass tube (D) at (F). Continuous flow is allowed by connecting one of the branches of tube (D) to a glass tube which extends about 20 cm. beneath the surface of water in a bottle (G) fitted with a two-hole stopper. The air flow is regulated at the outlet of the laboratory line of compressed air and by a screw clamp (H) on a short piece of tubing connected to the free arm of a "Y" tube interposed in the pressure line. The maximum pressure obtainable in the system is regulated by the depth of water in the pressure bottle (G), and as it is in the range of 18 to 20 cm. of water, small-caliber, thin-walled, soft rubber tubing may be used for the connections. The rate of delivery from the capillary micropipette is controlled by varying the degree of closure of tube (I) which is attached to the remaining arm of tube (D) and has a mouth piece at its distal end.

Rinsing the pipette thirty times with the suspension to be studied and then filling to the zero mark is accomplished rapidly by pinching closed the two rubber tubes of the pressure system at (J) and alternately applying suction and pressure through the mouth piece tube (I). Similarly, the pipette is cleaned with concentrated nitric acid, distilled water, alcohol and ether, using a short rubber tube with a mouth piece.

For delivery, the pressure in the system is adjusted by means of the screw clamp at (H) and the depth of water in the pressure bottle (G), so that when the pipette tip is not in contact with a body of fluid or a surface, no movement of the fluid in the pipette occurs when the mouth piece tube (I) is pinched closed. When a surface is touched to the pipette tip, closure of the mouth piece tube causes the capillary micropipette to empty at the desired speed. A magnifying glass, held in position by a clamp attached to the ring stand, is used to visualize the meniscus.

Below the pipette a rack and pinion are used to raise a glass slide to contact the pipette tip. The surface of the slide must be perpendicular to the vertical axis of the orifice of the pipette tip and the contact not so firm as to prevent flow. For this purpose the slide may be placed across the upper end of the draw tube of a microscope, or a mechanical

stage can be modified and held in place by a clamp attached to the ring stand, permitting a fine making and breaking of the contact.

Droplets of suspensions containing 0.08 to 0.2 per cent formalin delivered upon glass slides coated with a thin film of serum (by distributing a small drop of 50 per cent serum in distilled water in the manner of making a blood smear) were found to form a distinct margin upon drying which clearly delineated the droplet area in the stained preparation under the microscope.

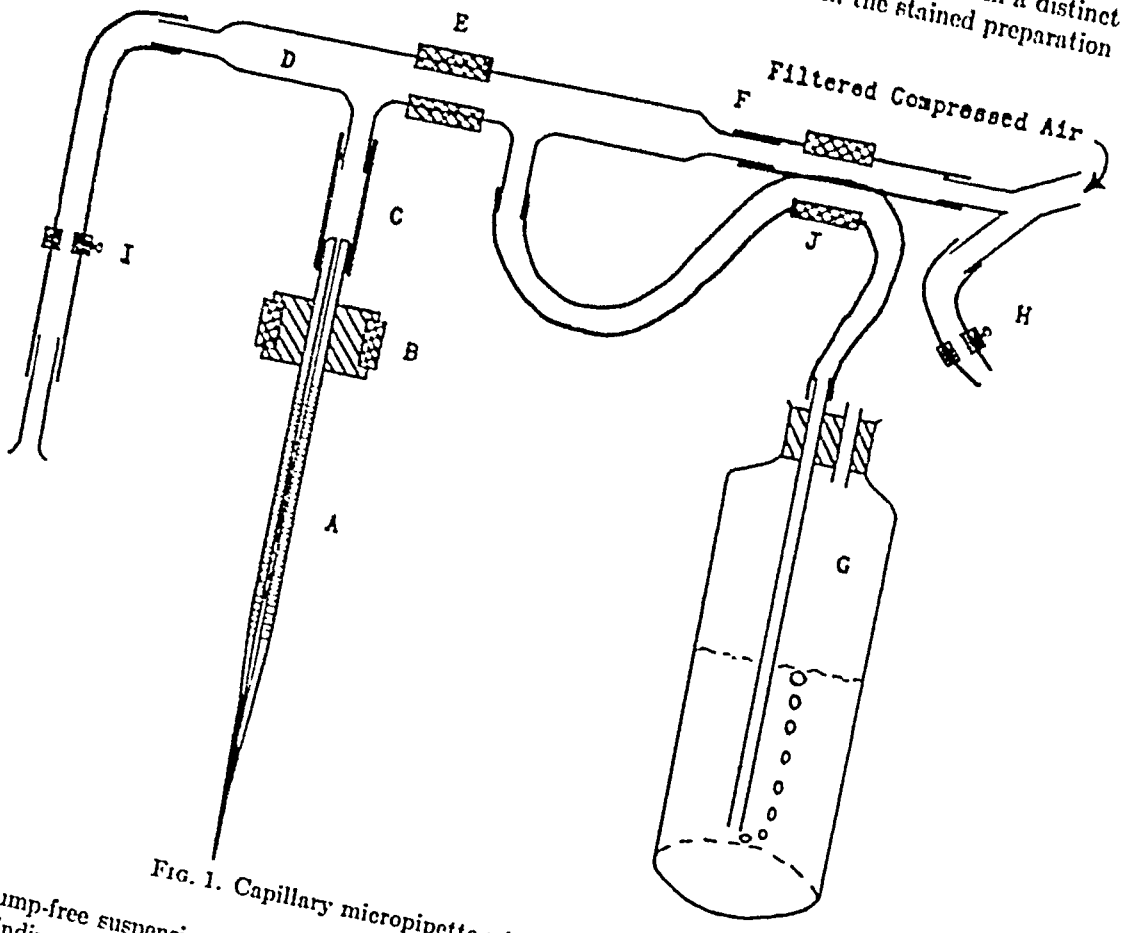

FIG. 1. Capillary micropipette with pressure system

Clump-free suspensions of tubercle bacilli containing only single cells were prepared by grinding a seven-day-old culture of H37-Rv in 0.2 per cent formalin, filtering through Whatmann # 42 filter paper twice, diluting to desired density and heating at 80°C. in a water bath for fifteen minutes. All suspensions were shaken mechanically for thirty minutes prior to use.

For counting, a Zeiss binocular microscope with 1.5× magnifying draw-tube prisms, achromatic 1.25 HI 90× oil immersion objective, and Abbe condensor was used. When counting pneumococci, 10× Huyghenian oculars were used (field area of 0.0094 mm.^{2}). When counting tubercle bacilli, 5× Huyghenian oculars were used (field area of 0.0276 mm.^{2}).

OBSERVATIONS

In order to determine the variability of aliquots from consecutive 5 mm. segments of the pipette as compared to the variability of aliquots delivered from the same segment of the pipette, a suspension of tubercle bacilli with a computed density of about 593,000 cells per cc. was used. Twelve aliquots of 0.23 cu. mm. size were delivered from consecutive segments of the pipette and twelve from the same segment of the pipette. It was noted that the former method was much more easily and rapidly accomplished than the latter. The droplets were delivered onto serum spread slides, air dried, heat fixed, and Ziehl-Neelsen stained. The variability of the counts is shown in table 2.

In order to determine the variability of aliquots of 0.23 cu. mm. size as compared with aliquots of 0.46 cu. mm. size, a suspension of tubercle bacilli with a

TABLE 2

Influence of mode of delivery, size of aliquot and error of counting on the variation of counts

EXPERIMENT	DENSITY OF SUSPENSION IN CELLS/CC.	NUMBER OF SAMPLES	MEAN COUNT	STANDARD DEVIATION OF MEAN (±)	COEFFICIENT OF VARIATION
					per cent
1. Consecutive segments	593,000	12	136.6	15.8	11.6
Single segment	593,000	12	149.5	21.5	14.4
2. 0.23 mm.^{3} aliquots	744,000	10	170.7	27.13	15.9
0.46 mm.^{3} aliquots	744,000	10	357.5	20.50	5.7
3. Duplicate counting:					
(a) Pneumococcus vaccine:					
First count	550,000	20	125.0	14.98	11.98
Second count	550,000	20	128.4	13.57	10.57
Error in counting = ±6.94 (5.48 per cent of the mean of the 40 counts)					
(b) Tubercle bacilli:					
First count	640,000	20	148.0	20.03	13.5
Second count	640,000	20	146.5	20.05	13.7
Error in counting = ±5.47 (3.71 per cent of the mean of the 40 counts)					

computed density of about 744,000 cells per cc. was used. Ten aliquots of 0.23 cu. mm. and ten aliquots of 0.46 cu. mm. size were studied. The larger aliquots showed a much smaller variability, as shown in table 2.

The error of counting was determined by the following procedures. Thirty-seven 0.23 cu. mm. aliquots of a pneumococcus vaccine, diluted 4,800 times in saline containing 0.15 per cent formalin, were delivered, air dried, gram stained and counted. The results showed a mean count of 126.5 organisms per aliquot with a standard deviation of ±15.16, *i.e.*, a coefficient of variation of 12 per cent. The density was computed to be about 550,000 cells per cc. in the diluted vaccine. Twenty of these aliquots were counted twice and the error of counting was determined as the square root of the sum of the squares of the differences between the duplicate counts divided by twice the number of pairs, *i.e.*, ±6.94 or 5.48 per cent of the mean of the forty counts (table 2).

Twenty 0.23 cu. mm. aliquots of a suspension of tubercle bacilli with a com-

puted density of about 640,000 cells per cc. were studied in like manner. The error in counting, estimated by the same procedure, was ±5.47 or 3.71 per cent of the mean of the forty counts (table 2).

A study was made of the range of variability as related to the density of the clump-free suspensions of tubercle bacilli in dilute formalin (0.08 to 0.2 per cent in distilled water). These counts were made on aliquots of 0.23 cu. mm. size and the densities were computed on the basis of the counts. As may be seen in table 3, very dilute suspensions with 75,000 tubercle bacilli per cc. or less showed a great variability in the bacterial content of the 0.23 cu. mm. aliquots. With suspensions containing from 350,000 to 850,000 tubercle bacilli per cc., the variability of the bacterial content was much less. In this range of density the maximum coefficient of variation of 15.9 per cent is much less than the 50 per cent error reported to be present with counting chamber determinations of suspensions containing less than 50 million cells per cc. Moreover, the range of the coefficient of variation from 8 to 16 per cent compares favorably with that

TABLE 3

Variation in counts as related to the density of the cell suspensions

DENSITY OF SUSPENSIONS IN CELLS/CC.	NUMBER OF 0.23 MM.³ SAMPLES	MEAN NO. CELLS PER 0.23 MM.³	STANDARD DEVIATION OF MEAN (±)	COEFFICIENT OF VARIATION
				per cent
14,000 to 40,000	12	6.25	2.93	46.9
34,000 to 75,000	10	12.45	4.68	37.6
374,000 to 435,000	12	93.00	6.99	7.53
362,000 to 477,000	12	95.75	12.5	13.05
526,000 to 661,000	12	136.6	15.84	11.6
548,000 to 730,000	20	148.0	20.03	13.5
626,000 to 861,000	10	170.7	27.13	15.9

found in Breed-smear counts of suspensions with from one to four million cells per cc.

SUMMARY

1. The principal direct methods of standardizing bacterial suspensions have been briefly discussed in regard to their limitations and degrees of precision.

2. A modification of the Linderstrom-Lang pipette has been described together with details of its operation and a method of using it to make small smear preparations of bacterial suspensions.

3. Experiments with a capillary micropipette having a uniform bore throughout its working length of 6 cm. indicate that delivery of twelve aliquots through successive 5 mm. segments can be accomplished with much greater speed than can be obtained by refilling the pipette to the same place and delivering from the same 5 mm. segment each time. Moreover, this method of consecutive deliveries did not increase the variability of the bacterial counts on these aliquots.

4. By counting all the bacteria in a stained droplet smear of the bacterial suspension, the errors involved in counting limited numbers of fields in stained smears with irregularly distributed organisms are avoided. Total counts of the organisms in a smear of a 0.23 cu. mm. aliquot require fifteen to twenty minutes with the optical system used. A microscopic factor of only 4,348 is used to convert the count to organisms per cc. of the suspension. If aliquots of 0.46 cu. mm. are used, thirty to forty-five minutes are required for counting, but the microscopic factor is reduced to 2,174. By counting ten of the 0.23 cu. mm. aliquots, with three and one half hours counting time, the factor may be reduced to 435.

5. The error of counting the organisms in a 0.23 cu. mm. aliquot was found to be 5.48 per cent of the mean count with smears containing about 130 pneumococci each; and 3.71 per cent with smears containing about 150 tubercle bacilli each.

6. Preliminary experiments with this method indicate that the coefficient of variation of counts of suspensions of tubercle bacilli containing between 350,000 and 850,000 organisms per cc. is less (8 to 16 per cent) than with suspensions containing 75,000 or less organisms per cc. (38 to 47 per cent). Moreover, the coefficient of variation of these counts is less when aliquots of 0.46 cu. mm. are used (5.7 per cent) than when aliquots of 0.23 cu. mm. are used (15.9 per cent).

CONCLUSIONS

The direct microscopic counting of organisms in droplet-size stained smears of known small volumes of suspensions delivered by means of a capillary micropipette is presented as a method of standardizing dilute suspensions of bacteria.

The method may be compared favorably with existing methods as to simplicity, inexpensiveness, speed and precision.

Preliminary studies indicate that this method may be used to standardize dilute suspensions of bacteria with a precision at least equal to that of the Breed-smear technique and greater than that of the chamber-counting methods.

SUMARIO

Técnica Directa para Estandardizar por medio de una Micropipeta Capilar las Suspensiones Diluídas de Bacilos Tuberculosos

1. Analízanse sucintamente, con respecto a sus limitaciones y precisión, las principales técnicas directas para normalizar las suspensiones bacterianas.

2. Descríbese una modificación de la pipeta de Linderstrom-Lang, junto con pormenores relativos a su empleo y un método para usarla en la preparación de pequeños frotes de suspensiones bacterianas.

3. Los experimentos realizados con una micropipeta capilar de un calibre uniforme de 6 cm. en todo su largo efectivo indican que puede obtenerse la entrega de doce partes alícuotas a través de segmentos sucesivos de 5 mm. con una velocidad mucho mayor que la obtenible rellenando la pipeta hasta la misma marca y vertiendo cada vez del mismo segmento de 5 mm. Además, esta técnica

de entregas consecutivas no acrecentó la variabilidad de las numeraciones bacterianas en dichas partes alícuotas.

4. Contando todas las bacterias en un frote en gotilla teñido de la suspensión bacteriana, evítanse los errores derivados de la cuenta de cantidades limitadas de campos en fotes teñidos y en que los microbios están distribuídos desigualmente. En un frote de una parte alícuota de 0.23 mm.^{3}, la numeración total de los microbios consume de 15 a 20 minutos con el sistema óptico utilizado. Para convertir la numeración a gérmenes por cc. de suspensión, se emplea un factor microscópico de no más de 4,348. Si se usan alícuotas de 0.46 mm.^{3}, se necesitan de 30 a 45 minutos para la numeración, pero el factor microscópico desciende a 2,174, y contando 10 de las alícuotas de 0.23 mm.^{3}, con tres horas y media de numeración, puede bajar a 435.

5. El error cometido al contar los gérmenes en una parte alícuota de 0.23 mm.^{3} resultó ser 5.48 por ciento de la fórmula media en los frotes que contenían unos 130 neumococos cada uno; y 3.71 por ciento en los que contenían unos 150 bacilos tuberculosos cada uno.

6. Los experimentos preliminares verificados con esta técnica indican que el coeficiente de variación en las numeraciones es menor (8 a 16 por ciento) en las suspensiones que contienen entre 350,000 y 850,000 bacilos tuberculosos por cc. que en las que contienen 75,000 o menos por cc. (38 a 47 por ciento). Además, el coeficiente de variación es menor cuando se emplean partes alícuotas de 0.46 mm.^{3} (5.7 por ciento) que con alícuotas de 0.23 mm.^{3} (15.9 por ciento).

CONCLUSIONES

La numeración microscópica directa de los gérmenes en los frotes de tamaño de gotillas teñidos de pequeños volúmenes de suspensiones entregadas por medio de una micropipeta capilar se presenta como técnica apropiada para estandardizar las suspensiones diluídas de bacterias.

Esta técnica compárase favorablemente con las técnicas actuales en cuanto a sencillez, baratura, celeridad y precisión.

Los estudios preliminares indican que puede utilizarse esta técnica para normalizar suspensiones diluídas de bacterias con una precisión por lo menos igual a la técnica de frotes de Breed y mayor que la de las técnicas bacteriométricas.

Acknowledgment

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FACTORS AFFECTING THE SENSITIVITY IN VITRO OF TUBERCLE BACILLI TO STREPTOMYCIN[1,2,3]

Nov. 1949 march.

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INTRODUCTION

The use of streptomycin as an adjunct in the treatment of tuberculosis and the emergence of strains of tubercle bacilli resistant to the drug in the patient raise the question of adequate laboratory tests for the detection of these resistant strains. The Dubos and Davis liquid Tween albumin medium (1) has been reported by Wolinsky and Steenken (2) as an "ideal one for determining streptomycin sensitivity of virulent human or bovine tubercle bacilli." Smith (3) has suggested that turbidimetric measurements in Tween albumin medium provide an accurate method for measuring growth of *M. tuberculosis* in the presence of streptomycin. Fisher (4), however, first called attention to the marked discrepancy between the sensitivity to streptomycin of tubercle bacilli tested in Tween albumin medium and the sensitivity of bacilli tested in the modified Proskauer and Beck medium enriched with serum, introduced by Youmans (5) and by Youmans and Karlson (6).

Fisher (7) found that Tween 80 alone was responsible for as much as a thousandfold increase in the inhibitory power of streptomycin for tubercle bacilli. Glycerol exerted a similar action but to a lesser degree. Fisher recommended that a test medium for streptomycin sensitivity should consist only of a liquid synthetic medium enriched with plasma or serum.

In view of the importance of detecting resistance to streptomycin of *M. tuberculosis* and the lack of unanimity as to method among workers in the field, the following studies were undertaken.

MATERIALS AND METHODS

The Tween albumin medium (1) as used by Steenken (8) has the following composition.

KH_2PO_4	1.00 Gm.
Na_2HPO_4—12 H_2O	6.25 Gm.
Sodium citrate—2 H_2O	1.50 Gm.
$MgSO_4$—7 H_2O	0.60 Gm.
Glucose	2.00 Gm.
Asparagin	2.00 Gm.
Distilled H_2O	1,000.00 ml.
pH 7.00	

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[1] From the Department of Bacteriology, Northwestern University Medical School, Chicago, Illinois.

[2] Presented in part at the 44th Annual Meeting of the National Tuberculosis Association, New York, New York, June 16, 1948.

[3] This investigation was supported in part by a research grant from the Division of Research Grants and Fellowships of the National Institute of Health, U. S. Public Health Service.

These are autoclaved at 10 lbs. for 20 minutes before the addition of:

Vegex	0.2 Gm.
Tween 80	0.5 Gm.

The Vegex and the Tween 80 are autoclaved separately in 10.0 per cent aqueous solution at 10 lbs. for twenty minutes, and are added aseptically to the medium. Finally, bovine albumin (Fraction V) is mixed with saline to form a 10 per cent solution, inactivated at 55° for thirty minutes, sterilized by Berkefeld filtration, and then added aseptically to the above medium to give a final concentration of 0.2 per cent.

The serum-synthetic medium (6) (Modified Proskauer and Beck) is composed of the following ingredients:

Asparagin	5.0 Gm.
KH_2PO_4	5.0 Gm.
K_2SO_4	0.5 Gm.
Glycerol	20.0 Gm.
Distilled H_2O	1,000.0 ml.

The pH is adusted to 7.0 with 40 per cent sodium hydroxide and then magnesium citrate, 1.5 Gm., is added.

The solution is autoclaved at 15 lbs. for twenty minutes and sufficient filtered sterile serum (human, bovine, or horse) is added to make a final concentration of 10 per cent.

The two media were dispensed respectively in 100 or 200 ml. volumes to fourteen flasks where the appropriate amounts of streptomycin were added to all but one flask. The streptomycin (streptomycin hydrochloride) was dissolved in 0.01 molar phosphate buffer pH 7.0 and so added to make a final concentration in the flasks of media of: 1,000, 500, 100, 50, 25, 12.5, 6.25, 3.125, 1.56, 0.78, 0.39, 0.195, and 0.095 γ per ml. Finally, the media were dispensed in 5 cc. amounts to Pyrex test tubes (20 by 150 mm.) and capped with aluminum caps. These tubes could be kept in the refrigerator four or five weeks without loss of streptomycin potency.

The bovine alubmin (Fraction V) used throughout was obtained from Armour Laboratories (Lots C1403 and C1765).

Tween 80 (Lot 5669) was obtained from the Atlas Powder Company from a lot recommended for bacteriological use.

OBSERVATIONS

Comparison of Tween Albumin Medium With Serum-synthetic Medium

The sensitivity to streptomycin of the cultures of tubercle bacilli was determined by inoculating equal amounts of the cultures into the thirteen streptomycin concentrations and the control. The inocula used consisted of either 0.1 or 0.25 mg. wet weight of either a ground suspension or a seven day culture in Tween albumin medium. The same kind and amount of inoculum was employed in each case where comparisons were made. The tubes were incubated for twenty-eight days and examined at seven day intervals. The sensitivity to streptomycin of the culture was defined as the least amount of streptomycin which completely inhibited growth.

Most strains of *M. tuberculosis* grew so rapidly in both of the above media that readings could be made at the end of seven days. With some cultures, growth appeared upon further incubation in some of the concentrations of streptomycin in which growth had not been visible earlier.

In table 1 may be seen the sensitivity to streptomycin of 66 recently isolated strains of tubercle bacilli at the end of fourteen days of incubation. A twofold difference in sensitivity between strains was considered to be within the limits

TABLE 1

A comparison of the sensitivity to streptomycin of the 66 strains of tubercle bacilli on serum-synthetic medium and Tween albumin medium

SENSITIVITY TO STREPTOMYCIN AFTER FOURTEEN DAYS OF INCUBATION*					
Strain Number	Serum-synthetic Medium	Tween Albumin Medium	Strain Number	Serum-synthetic Medium	Tween Albumin Medium
129	>1,000	>1,000	180	50	12.5
112	>1,000	>1,000	191	12.5	0.78
175	>1,000	>1,000	173	1.56	0.39
178	>1,000	50	176	6.25	0.78
195	>1,000	6.25	188	6.25	0.095
103	>1,000	>1,000	184	3.12	0.19
106	>1,000	>1,000	185	3.12	0.19
111R	>1,000	100	198	3.12	0.39
197	>1,000	>1,000	207	3.12	0.095
200	>1,000	>1,000	160	1.56	0.19
201	>1,000	>1,000	189	1.56	0.095
202	>1,000	>1,000	196	1.56	0.19
203	>1,000	>1,000	15	1.56	0.095
205	>1,000	>1,000	206	1.56	0.19
208	>1,000	>1,000	211	1.56	0.39
210	>1,000	>1,000	212	1.56	0.39
177	1,000	500	165	0.78	0.19
192	1,000	25	166	0.78	0.19
193	1,000	25	167	0.78	0.19
95	1,000	25	168	0.78	0.19
23R	1,000	50	169	0.78	0.19
187	100	3.125	170	0.78	0.19
194	500	25	171	0.78	0.19
58	500	25	172	0.78	0.19
73	500	12.5	174	0.78	0.19
128	500	25	186	0.78	0.095
24R	500	12.5	H37Rv	0.78	0.19
69R	500	25	6	0.78	0.095
100R	500	12.5	209	0.78	0.095
121	500	25	107	0.39	0.19
97R	500	6.25	182	0.39	0.19
204	500	12.5	183	1.56	0.19
213	500	50	190	0.39	0.095

* In γ per ml.

of error of the method. Fifty of the 66 cultures tested showed a resistance to streptomycin 4 to 160 times greater in the serum-synthetic medium than in the Tween albumin medium. Similar differences were observed after twenty-eight days of incubation. This discrepancy is similar to that reported by Fisher (4, 7)

and was attributed by him to the enhancement of the action of streptomycin by Tween 80.

Comparable streptomycin sensitivities in the two media were observed for 16 of the 66 strains. In both media, 13 of these 16 strains had a sensitivity to streptomycin greater than 1,000 γ per ml., the maximum amount of streptomycin used in the tests. If tubes had been used containing higher concentrations of streptomycin, it is possible that a discrepancy between the results of the streptomycin sensitivity tests with these organisms in the two media might have been revealed.

Before exploring the reason for the differences observed, however, it seemed advisable to investigate the role played by the size and type of inoculum employed in the tests.

Inoculum

The type of inoculum proposed (2) for streptomycin sensitivity tests in Tween albumin medium is a culture of tubercle bacilli grown in this medium for two seven day culture generations in order to ensure a well dispersed culture. The inoculum is then standardized by turbidimetric methods.

The inoculum originally proposed for the serum-synthetic medium is prepared by mechanically grinding the growth of tubercle bacilli obtained from solid media (6); determining the wet weight of cells per ml. by centrifugation in a Hopkins vaccine tube; and finally, diluting with 0.01 molar phosphate buffer to obtain the desired concentration of cells.

In the present study, the amount of growth in a seven day old culture of Tween albumin medium was also determined by centrifugation in Hopkins vaccine tubes. If this procedure indicated the presence of more tubercle bacilli than desired for any given test, the appropriate dilution was made with Tween albumin medium. If the culture had too few cells, it was incubated one or two days longer, and then restandardized.

The following tests were carried out to determine the effect of the nature of the inoculum, using 8 strains of tubercle bacilli, and of the amount of the inoculum, using 10 strains of tubercle bacilli.

Bacterial suspensions were prepared from Herrold's (9) egg yolk agar medium cultures and 0.25 mg. of each was suspended in 0.2 ml. of buffer solution. The suspensions were then delivered to each of the thirteen tubes of serum-synthetic medium forming the series of streptomycin concentrations, and to the controls. The Tween albumin cultures were standardized, and 0.25 mg. of these cells in 0.1 ml. Tween albumin medium was delivered to each of the thirteen tubes of serum-synthetic medium, and to the controls. After both sets of cultures had been incubated for fourteen days, the sensitivity to streptomycin obtained with the two types of inoculum was found to be the same. The tests were repeated with the same two types of inoculum, but with Tween albumin as the medium for the sensitivity test. In these tests also the streptomycin sensitivities were not affected by the kind of inoculum used. It should be mentioned that diffuse growth was obtained in the Tween albumin medium when a suspension prepared

by grinding was used as an inoculum as well as when suspensions obtained from growth in the Tween albumin medium were used as inocula.

Therefore, whatever ingredients might be responsible for the discrepancy previously observed between the streptomycin sensitivities in the two media, they do not occur in sufficient amount in the menstrua used for the inocula to affect the results.

One of the writers has previously reported (5) that within certain limits the size of the inoculum does not alter significantly the streptomycin sensitivity of the H37Rv strain in serum-synthetic medium. Further experiments on the effect of the size of inoculum were carried out as follows.

The inoculum was prepared from organisms grown in Tween albumin medium and separate tests were conducted with each of the 10 strains, using 0.25 mg., 0.1 mg. and 0.01 mg. of tubercle bacilli as inocula. Tween albumin and serum-synthetic media were inoculated.

With all strains, both the 0.25 mg. and 0.1 mg. inocula gave the same results in serum-synthetic medium. The results in Tween albumin medium using the above inocula were similar to each other although, as previously noted, they differed from those in serum-synthetic medium.

With an inoculum of 0.01 mg., however, the sensitivity to streptomycin of the cultures of tubercle bacilli was greater at fourteen days than with either the 0.25 mg. or the 0.1 mg. inoculum. After twenty-eight days of incubation, however, the streptomycin sensitivities were the same regardless of the size of the inoculum. This was true in both serum-synthetic and Tween albumin medium, except for a few strains which had a sensitivity to streptomycin greater than 1,000 γ per ml. These grew well in seven days in all tubes regardless of the size of the inoculum.

Within the limits set by the above experiments, the size or nature of the inoculum did not affect the sensitivity of tubercle bacilli to streptomycin.

Factors Responsible for the Discrepancy between Sensitivity of Tubercle Bacilli to Streptomycin in Tween Albumin and Serum-synthetic Media

Fisher (7) has reported that the discrepancy observed between the results of streptomycin sensitivity tests when carried out in Tween albumin medium and in serum-synthetic medium results primarily from the fact that Tween 80 increases the bacteriostatic effect of streptomycin. He also observed that glycerol had a similar action, but to a lesser degree.

As streptomycin assays of the two media using Romansky's method (10) showed that the streptomycin was not reduced in potency in either medium, the following studies were undertaken.

Six different modifications of the two media were prepared:

(S) Synthetic medium without glycerol or serum;
(SG) S + 2 per cent glycerol;
(SS) S + 10 per cent serum;
(SGS) S + 10 per cent serum + 2 per cent glycerol;
(DA) Dubos medium without Tween 80 but with 0.2 per cent albumin;
(DTA) DA + 0.05 per cent Tween 80

Eighteen strains of tubercle bacilli were used for testing the four modifications of the serum-synthetic medium. An inoculum (0.25 mg.) of each of the strains, prepared by grinding, was delivered respectively into tubes of each of the four media (S, SG, SS, SGS) containing the 13 concentrations of streptomycin previously employed. These tubes were incubated for twenty-eight days and examined every seven days.

In table 2 may be seen the sensitivity to streptomycin of each of these strains when grown for two weeks in the four modifications of the serum-synthetic medium.

TABLE 2

A comparison of the sensitivity to streptomycin of strains of human type tubercle bacilli when grown in various modifications of serum-synthetic medium and Tween albumin medium*

STRAIN NUMBER	TYPE OF MEDIUM					
	S	SG	SS	SGS	DA	DTA
165	0.39	0.39	0.78	0.78	0.78	0.19
173					3.125	0.39
H37Rv	0.78	1.56	1.56	1.56	1.56	0.39
H37RvR	>1,000.0	>1,000.0	>1,000.0	>1,000.0	>1,000.0	>1,000.0
200	50.0	1,000.0	1,000.0	1,000.0	500.0	1,000.0
204	25.0	100.0	500.0	500.0	500.0	50.0
207	0.78	0.39	3.125	6.25	1.56	0.39
192	500.0	1,000.0	1,000.0	1,000.0	500.0	50.0
178	500.0	500.0	1,000.0	1,000.0	1,000.0	50.0
196	N.G.	0.39	100.0	50.0	1.56	0.095
170	0.095	0.19	0.78	0.78		
182	0.19	0.39	0.78	1.56		
183	0.095	0.39	1.56	1.56		
209	0.095	0.095	0.39	0.39		
185	0.095	0.78	3.125	3.125		
167	0.095	0.39	0.78	0.78		
15	0.095	0.19	0.78	0.78		
188	N.G.	0.19	3.125	6.25		
212	N.G.	0.19	0.39	0.39		

N.G. = no growth in any tube.
* In γ per ml.

Effect of glycerol: Of the 18 strains tested, 10 after 14 days of incubation showed the same sensitivity to streptomycin in the basic synthetic medium with glycerol (SG) as in the medium without glycerol (S), (table 2). Five strains were more resistant in the presence of glycerol. No conclusions could be drawn concerning the 3 strains which failed to grow in fourteen days in the medium without glycerol.

The effect of glycerol in the presence of serum was determined by comparing the streptomycin sensitivities in the medium SGS with those in SS (table 2). The results were the same for all strains in both SGS and SS. Thus, the effect of glycerol alone, noted above, was not apparent in the presence of serum.

Effect of serum: The amount of growth of all strains in the control tubes was greater with serum than without. Three of the 18 strains did not grow in the basic synthetic medium without the serum after fourteen days of incubation. The effect of adding serum to the basic medium on the sensitivity of the remaining 15 strains to streptomycin may be seen by comparing SS with S in table 2. Ten strains were more resistant to streptomycin (4- to 32-fold) in the presence of serum. The remaining five gave similar results in the presence or absence of serum. The sensitivity to streptomycin of 10 strains was also decreased when serum was added to medium containing glycerol (SG compared with SGS, table 2).

Effect of Tween 80: Ten of the strains used for testing the ingredients of the serum-synthetic medium were also employed for the tests with the Tween albumin medium.

In table 2 may be seen the sensitivity to streptomycin of the ten strains in the Dubos medium plus albumin only (DA) and in the Dubos medium plus Tween and albumin (DTA).

The effect of Tween 80 on the inhibiting action of streptomycin is seen with 8 of the 10 strains and is lacking with only the 2 strains of a resistance greater than the streptomycin concentrations employed. The potentiation is from 4- to 20-fold with these particular strains. This is even more apparent if the results obtained with the 10 strains in SGS and DA are compared (table 2). With the exception of strain 196, the results in these two media are the same. Strain 196 did not give consistently similar results when sensitivity tests were repeated. This may be a reflection of the ease with which it produces streptomycin-resistant variants (11).

Although growth of the tubercle bacilli is frequently not as rapid, albumin can be used as a substitute for serum when preparing media for streptomycin sensitivity tests.

These results clearly confirm those of Fisher (7) that Tween 80 is the agent primarily responsible for the discrepancies observed between the results of streptomycin sensitivity tests in Tween albumin and serum-synthetic medium.

Effect of Amount of Tween 80 on the Bacteriostatic Action of Streptomycin

Batches of Tween albumin medium were prepared containing 0.05, 0.04, 0.03, 0.02, 0.01, 0.005, 0.003, 0.001 and 0.0005 per cent Tween 80, respectively, and streptomycin was incorporated in each in the same concentrations previously employed. Using 0.25 mg. of eight to ten day cultures of 6 strains of *M. tuberculosis* in Tween albumin medium as an inoculum, the nine series above were planted and incubated for fourteen days. As the inoculum itself was suspended in 0.1 ml. of 0.05 per cent Tween 80, the total Tween concentration in any one tube was increased by 0.001 per cent. Table 3 shows the sensitivity to streptomycin of the 6 strains grown in these media, and in serum-synthetic medium.

The cultures in the 0.05 per cent Tween 80 series of streptomycin dilutions were all well dispersed when the tubes were shaken after two weeks of incubation. In tubes with less Tween the degree of dispersion was less constant. For each

strain there was a critical concentration of Tween 80 below which the amount of dispersion observed in the Tween albumin medium containing 0.05 per cent Tween did not occur. The sensitivity to streptomycin also was not the same in all the concentrations of Tween 80. As the amount of Tween 80 was diminished, the sensitivity to streptomycin became progressively less. For each strain, however, there was a point where further decrease in the amount of Tween 80 produced no significant change in sensitivity to streptomycin. At this point, also, dispersion was no longer produced by the amount of Tween 80 present.

TABLE 3

The effect of the concentration of Tween 80 on the streptomycin sensitivity and dispersion of tubercle bacilli*

CULTURE NUMBER	SERUM MEDIUM	CONCENTRATION OF "TWEEN 80" IN PER CENT										
		0.051	0.041	0.031	0.021	0.011	0.006	0.005	0.004	0.002	0.0015	0.001
165 Streptomycin sensitivity	1.56	+ 0.095			+ 0.095	+ 0.095	+ 0.095	± 0.39	± 0.39	± 0.78	− 0.39	− 0.39
178 Streptomycin sensitivity	500	+ 50.0	+ 50.0	+ 50.0	+ 50.0	+ 50.0	+ 50.0	+ 500.0	± 500.0	− 500.0	− 500.0	− 1000
196 Streptomycin sensitivity	3.12	+ 0.095	+ 0.19	+ 0.19	+ 0.19	+ 0.19	+ 0.19		+ 0.39	± 0.39	− 0.78	− 0.78
187 Streptomycin sensitivity	100	+ 1.56	+ 1.56	+ 1.56	+ 3.12	+ 6.25	+ 6.25	+ 50.0	+ 50.0	± 50.0	± 50.0	− 50.0
204 Streptomycin sensitivity	500	+ 25.0	+ 25.0	+ 25.0	+ 25.0	+ 25.0	+ 50.0	+ 50.0	+ 100.0	− 100.0	− 100.0	− 100.0
173 Streptomycin sensitivity	1.56	+ 0.095	+ 0.095	+ 0.19	+ 0.19			+ 0.39	+ 0.39	± 0.39	− 0.78	− 0.78

+ = Dispersed growth
± = Partially dispersed growth
− = No dispersion
* In γ per ml.

Effect of Tween 80 on Nonacid-fast Organisms

In view of the fact that the previous tests indicated a relation between the degree of dispersion produced by Tween 80 and the potentiating effect on the bacteriostatic action of streptomycin, an investigation was undertaken to determine whether bacteria which do not owe their dispersal to Tween 80 would show an increase in sensitivity to streptomycin in the presence of Tween 80. Eleven strains of nonacid-fast bacteria were chosen which grew unclumped in broth. These were: 6 strains of *Staphyloccocus aureus*, 3 strains of *E. coli*, one of *E. typhi*, and one of *A. aerogenes*. Tubes of beef infusion broth containing streptomycin in concentrations varying from 40 γ per ml. to no γ per ml. (ten tubes) were prepared. A similar series was prepared, adding 0.05 per cent Tween

80 to the streptomycin broth. The inoculum for the streptomycin sensitivity tests with these organisms was 0.1 ml. of a 1 to 1 million dilution of a six hour culture of each strain. These streptomycin broths were inoculated with the test organisms, incubated, and inspected thereafter at twenty-four, forty-eight and seventy-two hours. The sensitivity to streptomycin of all strains was the same both in the presence and in the absence of Tween 80.

Modified Dubos Formula

McDermott and his associates (12) recommended the reduction of the Tween 80 to 0.02 per cent and the increase of albumin to 0.5 per cent in the Dubos formula to eliminate the potentiating effect of Tween 80. In the present studies this medium has been employed to test the sensitivity to streptomycin of 54 strains of tubercle bacilli, and the results were compared with those obtained in serum-synthetic medium. Of the 54 strains tested, 33 were four- to eightfold more resistant in the serum-synthetic media than in the modified Tween albumin medium. Of the 21 which gave essentially the same sensitivity in the two media, ten showed in both media a streptomycin sensitivity greater than 1,000 γ per ml. in both media. If the series had included concentrations of streptomycin greater than 1,000 γ per ml., it is possible that with these strains also a discrepancy between the results in the two media might have been revealed.

Dehydrated Media

Two other types of liquid media which were tested were those dispensed in dehydrated form (L14725 and L54720) by Difco. These media contained the salts as described by Dubos and Davis (1) plus proteose peptone No. 3, Bacto yeast extract and Bacto casitone. The only difference between the two media was that one (L14725) contained 0.05 per cent Tween 80. These Difco bases were dissolved in distilled water, and autoclaved in 90 cc. amounts. To this broth were added 10 cc. of sterile serum, containing 7.5 per cent dextrose, also supplied by Difco. Streptomycin sufficient to give the concentrations previously employed was added to the two media.

The sensitivity to streptomycin of 7 strains of tubercle bacilli was determined in the two Difco media and in serum-synthetic medium. The Difco medium, without Tween 80, gave results which were approximately the same as in the serum-synthetic medium. In the Difco medium containing Tween 80, however, there was a fourfold increase in the bacteriostatic action of streptomycin on 4 of the 7 strains tested.

Solid Media

Pyle (13) called attention to the fact that cultures of tubercle bacilli were not homogeneous with regard to their sensitivity to streptomycin. Using plates of Herrold's (9) glycerol egg agar medium containing different concentrations of streptomycin, she cultured *M. tuberculosis* from the sputum of patients who were to receive treatment with streptomycin. Pyle found that these cultures of tubercle bacilli were composed of cells which did not have a uniform sensitivity

to streptomycin. The same composite character existed in cultures taken during and after streptomycin treatment.

A sensitivity test that would give some indication of the relative numbers of streptomycin sensitive and resistant organisms in a culture would be of great value. Liquid media, however, are unsuitable for this purpose. A solid medium would permit an estimation of the relative amount of growth on the different concentrations of streptomycin. Moreover, such a medium might also be employed for the primary isolation and testing for sensitivity of tubercle bacilli to streptomycin in one process, thus reducing the time needed for the completion of the tests.

Herrold's Medium: Herrold's (9) glycerol-egg agar medium was prepared by autoclaving the nutrient agar base for thirty minutes in 150 ml. amounts and then adding a single egg yolk in a sterile manner after the agar had melted and been cooled to 50 C.

Basic agar:	Beef extract	3.0 Gm.
	Bacto Peptone	10.0 Gm.
	NaCl	50.0 Gm.
	Distilled H_2O	1,000.0 ml.
	Agar	15.0 Gm.
	Glycerol	60.0 ml.
	pH 7.2	

The appropriate amount of streptomycin was added to each flask just after the addition of the egg yolk. Twelve to fifteen ml. amounts were then converted into slants in screw-top test tubes and allowed to harden.

The inoculum for this series was prepared by growing cultures in Dubos medium for seven days, and then delivering 0.1 ml. (0.1 mg.) of this culture to each of the fourteen slants of Herrold's medium containing different concentrations of streptomycin. The inoculum was allowed to flow over the slants, which were then incubated in a horizontal position. At the end of seven days, the growth on the egg slant was not abundant enough to make accurate readings possible. After fourteen days of incubation, most strains showed profuse growth on the tube with no streptomycin and on some of the lower concentrations of streptomycin.

At this time (fourteen days) with streptomycin sensitive cultures, there was usually complete inhibition of growth in the higher concentrations of streptomycin. After four weeks of incubation, however, slight growth, consisting of one to several colonies, often appeared in one or two additional tubes containing concentrations of streptomycin two- to fourfold greater than the lowest concentration which had completely inhibited growth at fourteen days.

In table 4 may be seen the sensitivity to streptomycin of the 95 strains tested on Herrold's egg medium as compared with the results obtained with serum-synthetic liquid medium. Seventy-five of these strains (77.7 per cent) were found to be equally sensitive on both media at the end of two weeks, *i.e.* there was no more than a twofold difference in the results, the error inherent in the method. It should be noted, however, that this slight discrepancy was consistently in the

TABLE 4

Comparison of results of streptomycin sensitivity tests in serum-synthetic medium and Herrold's glycerol egg medium

SENSITIVITY TO STREPTOMYCIN AFTER FOURTEEN DAYS OF INCUBATION*					
Strain Number	Serum-synthetic Medium	Herrold's Medium	Strain Number	Serum-synthetic Medium	Herrold's Medium
15	1.56	3.125	333	>1,000.0	>1,000.0
69R	100.0	100.0	334	25.0	25.0
129	500.0	>1,000.0	335	>1,000.0	>1,000.0
165	0.39	0.78	336	6.25	12.5
170	0.78	1.56	337	>1,000.0	>1,000.0
181	100.0	25.0	338	100.0	50.0
182	0.78	1.56	339	>1,000.0	>1,000.0
183	1.56	3.125	341	500.0	500.0
187	100.0	50.0	342	>1,000.0	>1,000.0
188	0.78	3.125	343	50.0	50.0
189	0.78	0.78	344	1.56	1.56
196	0.78	3.125	345	1.56	3.125
197	>1,000.0	>1,000.0	346	1.56	6.25
200	>1,000.0	>1,000.0	347	0.78	1.56
202	>1,000.0	>1,000.0	348	1.56	3.12
204	500.0	1,000.0	349	1.56	3.12
207	3.125	6.25	350	0.78	1.56
208	>1,000.0	>1,000.0	351	1.56	3.125
209	0.78	3.125	354	0.78	3.125
210	>1,000.0	>1,000.0	355	0.78	1.56
211	0.19	0.78	356	0.39	1.56
214	3.125	3.125	363	1.56	3.125
223	1.56	3.125	366	3.125	6.25
225	0.78	1.56	373	1.56	6.25
228	3.125	25.0	394	>1,000.0	>1,000.0
230	500.0	500.0	395	1.56	1.56
231	0.78	3.125	396	1.56	3.125
236A	500.0	500.0	398	6.25	6.25
244	0.78	1.56	399	1.56	3.125
245	25.0	25.0	402	3.125	6.25
247	100.0	1,000.0	403	500.0	500.0
249	12.5	12.5	404	12.5	25.
254	6.25	6.25	405	>1,000.0	>1,000.0
260	3.125	3.125	406	1.56	12.5
263	0.78	3.125	407	12.5	50.
269	3.125	3.125	408	500.0	500.0
273	.39	1.56	409	3.125	6.25
274	3.125	0.78	411	>1,000.0	>1,000.0
294	1.56	3.125	414	1.56	6.25
299	1.56	6.25	422	0.78	3.125
314	>1,000.0	>1,000.0	425	500.0	500.0
322	1.56	3.125	430	1.56	6.25
323	6.25	12.5	438	1.56	3.125
324	1.56	3.125	439	500.0	100.0
325	1.56	3.125	442	25.0	25.0
326	1.56	3.125	477	>1,000.0	>1,000.0
327	6.25	3.125	478	>1,000.0	>1,000.0
332	3.12	1.56			

* In γ per ml.

direction of a greater resistance on the egg slants. At the end of twenty-eight days of incubation, 78 strains (80.2 per cent) were found to be equally sensitive in the two media. Only 2 of the 95 strains, however, showed more than a fourfold difference in sensitivity on the two media at the end of four weeks of incubation.

Serum-synthetic agar medium: The addition of 1.5 per cent agar to serum-synthetic-streptomycin medium may also be used for determining streptomycin sensitivity. Fifteen strains were tested in this medium and the values obtained were compared with the results in serum-synthetic liquid medium. The results were the same in both media, but growth was somewhat slower in the agar medium.

A third solid medium was prepared by adding agar to the synthetic medium without the addition of serum. The tubercle bacilli grew very poorly, however, and readings could seldom be made until the tubes had been incubated four weeks.

Of the three solid media tested, the Herrold medium had the fewest disadvantages. The serum-synthetic agar required the addition of fresh serum, a tedious process. The synthetic broth plus agar was not sufficiently nutritious to provide enough growth to be read easily.

Detection of Small Numbers of Streptomycin Resistant Tubercle Bacilli

As the usefulness of a streptomycin sensitivity test may depend on the ability of the medium employed to detect the presence of a small number of resistant organisms in any culture isolated from a patient, the use of mixtures of sensitive and resistant organisms as inocula for streptomycin sensitivity tests was studied. H37Rv (streptomycin sensitive) and H37RvR (resistant to more than 1,000 γ of streptomycin per ml.) were each grown in Dubos medium (0.05 per cent Tween) for one week, at which time 0.1 ml. of each culture contained approximately 0.1 mg. wet weight of the bacterial cells. The cultures were shaken to give the maximum degree of dispersion. The resistant culture was diluted decimally with Tween albumin medium so that it could be combined in very small amounts with a fixed amount of the sensitive strain and used as inocula for streptomycin sensitivity tests.

In these mixed cultures it was observed that the amount and the time of appearance of growth in the concentrations of streptomycin which completely inhibited growth of streptomycin-sensitive tubercle bacilli was proportional to the number of resistant organisms in the mixture.

In table 5 may be seen the sensitivities to streptomycin of 6 such mixtures containing different relative numbers of sensitive and resistant organisms and grown in modified Tween albumin, in serum-synthetic and in Herrold's egg medium. As may be seen in the table, the presence of a very small number of resistant organisms in the inoculum influenced the results markedly. When as little as 0.001 per cent (10^{-6} mg.) of the total inoculum was resistant to streptomycin, growth occurred in all concentrations of streptomycin in all media.

The amount of growth in the tubes of Herrold's medium containing sufficient

streptomycin to inhibit growth of the sensitive organisms was, within certain limits, proportional to the number of streptomycin resistant organisms present. With 0.001 per cent (10^{-6} mg.) of streptomycin-resistant organisms in the mixture, only 2 to 11 colonies were present on each slant of Herrold's medium at the end of fourteen days. With larger numbers of resistant bacilli, the growth was more confluent but much less in amount than on the control medium. A difference could be detected in the amount of growth with as many as, but not more than, 1.0 per cent (10^{-3} mg.) of streptomycin-resistant tubercle bacilli in the mixture. These differences, however, tended to disappear as the time of incubation was extended beyond fourteen days.

In the liquid media these differences in the amount of growth were far more difficult to detect.

TABLE 5

Result of streptomycin sensitivity tests on mixtures of sensitive and resistant tubercle bacilli after 28 days of incubation

MG. INOCULUM		STREPTOMYCIN SENSITIVITY OF MIXTURE*		
H37Rv	H37RvR	Serum-synthetic Medium	Modified Tween Albumin Medium	Herrold's Egg Medium
0	10^{-1}	>1,000.0	>1,000.0	>1,000.0
10^{-1}	0	1.56	0.19	1.56
10^{-1}	10^{-2}	>1,000.0	>1,000.0	>1,000.0
10^{-1}	10^{-3}	>1,000.0	>1,000.0	>1,000.0
10^{-1}	10^{-4}	>1,000.0	>1,000.0	>1,000.0
10^{-1}	10^{-5}	>1,000.0	>1,000.0	>1,000.0
10^{-1}	10^{-6}	>1,000.0	>1,000.0	>1,000.0
10^{-1}	10^{-7}	1.56	0.19	1.56
10^{-1}	10^{-8}	1.56	0.19	

* In γ per ml.

The most important conclusion to be drawn from these experiments is that cultures of tubercle bacilli may contain very few resistant organisms but under the conditions of the *in vitro* sensitivity tests may appear to be entirely composed of resistant tubercle bacilli.

DISCUSSION

The results of the present studies show that the sensitivity of cultures of tubercle bacilli to streptomycin *in vitro* is partially dependent on the composition of the culture medium employed. It has been reported by other investigators, Alture-Werber and Loewe (14), Wallace, Rhymer, Gibson and Shattuck (15), Green and Waksman (16), Donnovick and Rake (17), Lenert and Hobby (18), Hobby and Lenert (19) and Berkman, Henry and Housewright (20), that the sensitivity to streptomycin *in vitro* of other species of bacteria is also influenced by the composition of the culture medium. A variety of substances, including glucose, peptones, and serum tend to reduce the bacteriostatic action of streptomycin. While the mode of action of these agents is not clear, they may inter-

fere in some manner with the bacteriostatic action of streptomycin. In the present experiments it is of interest that the two agents, glycerol and serum, which decreased the sensitivity of the tubercle bacilli to streptomycin, also stimulated the rate of growth. It should be pointed out that not all of the strains of tubercle bacilli employed showed a significant decrease in sensitivity to streptomycin in the presence of glycerol and serum, although growth was apparently stimulated. In a previous report (5) it was observed that the sensitivity of tubercle bacilli to streptomycin was not appreciably affected by the presence of serum in the medium. Only one strain (H37Rv) was tested in the presence of serum in that study and the streptomycin sensitivity of this strain is not significantly affected by serum.

Tween 80, on the other hand, as shown by Fisher (4), increased the bacteriostatic action of streptomycin for tubercle bacilli and this effect was proportional to the concentration of Tween 80 in the medium. Tween 80 has a similar potentiating effect on the tuberculostatic action of subtilin (21), chloromycetin, certain synthetic organic compounds (22) and penicillin (23). The mechanism whereby Tween 80 increases the bacteriostatic action of streptomycin is not apparent from the data of the present study. It appears to be related to the surface active properties of the compound and may, as proposed by Kirby and Dubos (23), permit a more intimate contact between the antibiotic and the bacterial cell. It is of interest that Tween 80 did not increase the bacteriostatic action of streptomycin for nonacid-fast organisms, nor was it bacteriostatic for these organisms as it has been shown to be for tubercle bacilli (24).

The variability in the sensitivity to streptomycin of tubercle bacilli under different conditions emphasizes the necessity for care in the selection of a medium for streptomycin sensitivity tests. The ideal medium would be one which duplicated precisely the conditions which obtain *in vivo*. The original Tween albumin medium containing 0.05 Tween 80 and 0.2 per cent bovine albumin would appear to be unsuitable because of the marked potentiation of the bacteriostatic action of streptomycin by this concentration of Tween 80. The modification used by McDermott *et al.* (12), and by Sadusk and Swift (25), in which the concentration of Tween 80 is reduced to 0.02 per cent and the bovine albumin is increased to 0.5 per cent, was more suitable as the increase in the bacteriostatic action of streptomycin is no more than four- to eightfold. The serum-synthetic, bovine albumin synthetic, and Difco dehydrated liquid media are free of Tween 80, and such of these media as contain serum possibly simulate more closely the conditions present *in vivo*.

All liquid media, however, possess other disadvantages. They do not readily permit the detection of streptomycin resistant variant cells (11), nor the determination of the relative numbers of streptomycin sensitive and resistant organisms present in a culture of tubercle bacilli. Furthermore, liquid media are not as suitable for the determination of the sensitivity of tubercle bacilli to streptomycin at the time of primary isolation from the patient.

A solid medium in which streptomycin could be incorporated would solve at least partially the above objections to the use of liquid media. Extensive use of

Herrold's glycerol egg medium for the determination of the sensitivity of cultures of tubercle bacilli to streptomycin has shown it to be useful for this purpose, and Pyle (13) has employed this medium for the determination of the sensitivity to streptomycin of tubercle bacilli directly at the time of primary isolation from sputum. Other solid media may be found which may be even more suitable.

SUMMARY

A variety of media, both liquid and solid, have been tested for their suitability for determining the sensitivity of cultures of tubercle bacilli to streptomycin. The effect of Tween 80, bovine serum, glycerol, and the amount of inoculum on the sensitivity of cultures of tubercle bacilli to streptomycin was also determined.

It was found that Tween 80, even in low concentration, increased the sensitivity of the majority of cultures to streptomycin. Serum (bovine) increased the rate of growth and decreased slightly the streptomycin sensitivity of some strains. Glycerol increased slightly the resistance of a few strains of tubercle bacilli to streptomycin. In the presence of serum, however, glycerol had no observable effect.

Within the limits tested (0.01 to 0.25 mg. wet weight) the amount of inoculum employed did not affect the sensitivity to streptomycin provided a minimum incubation period of twenty-eight days was used. Large inocula, 0.1 to 0.25 mg., permitted completion of tests within fourteen days.

The sensitivity to streptomycin tended to be slightly less on solid medium than on liquid serum-synthetic medium. Nevertheless, it was felt that a solid medium such as Herrold's glycerol egg yolk agar was satisfactory for such tests and possessed the additional advantage of supplying an approximation of the relative numbers of streptomycin resistant and sensitive tubercle bacilli.

SUMARIO

Factores que Afectan la Sensibilidad in Vitro de los Bacilos Tuberculosos a la Estreptomicina

Varios medios, tanto líquidos como sólidos, han sido comprobados en cuanto a su adaptabilidad para determinar la sensibilidad de los cultivos de bacilos tuberculosos a la estreptomicina. Tambien se determinó el efecto del "Tween 80," el suero bovino, el glicerol y la cantidad de inóculo sobre la sensibilidad de dichos cultivos a la estreptomicina.

Observóse que el "Tween 8," aun a concentraciones bajas, acrecentaba la sensibilidad la mayoría de los cultivos a la estreptomicina. El suero (bovino) acrecentó la velocidad del desarrollo y atenuó ligeramente la sensibilidad de algunas cepas a la estreptomicina. El glicerol realzó levemente la resistencia de unas pocas cepas a la estreptomicina, pero, en presencia de suero, no ejerció efecto observable.

Dentro de los límites ensayados (0.01 por 0.25 mg. de peso húmedo), la cantidad de inóculo empleada no afectó la sensibilidad a la estreptomicina, con tal que se usara un período mínimo de incubación de veintiocho días. Los inóculos grandes (0.1 a 0.25 mg.) permitían completar las pruebas en término de catorce días.

La sensibilidad a la estreptomicina solió ser levemente menor con los medios sólidos que con los sero-sintéticos líquidos. Sin embargo, parece que un medio sólido, tal como el de agar-yema de huevo-glicerol de Herrold, resulta satisfactorio para dichas pruebas, poseyendo además la ventaja de revelar aproximadamente las proporciones relativas de bacilos tuberculosos resistentes y sensibles a la estreptomicina.

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THE

AMERICAN REVIEW OF TUBERCULOSIS ABSTRACTS

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Timetable of Tuberculosis.—Tuberculous infection in a person passes through a number of characteristic periods; in each of these periods a specific tuberculous condition appears frequently. The risk of falling ill with each specific type diminishes rapidly after the period characteristic of the condition. Stage 1 appears five to six weeks after the infection. It is characterized by tuberculin sensitivity, initial fever, erythema nodosum and the primary complex. There is nothing characteristic about the initial fever and only a tuberculin test indicates the true etiology of this, as well as of erythema nodosum. The sedimentation rate becomes elevated. Stage 2 occupies approximately the next three months. During this period, the malignant generalized forms of tuberculosis, particularly miliary and meningeal, occur. Sometimes, by means of special methods, tubercle bacilli may be cultured from the urine. Stage 3, the pleurisy period, follows immediately after Stage 2 and lasts about four months. The effusion is usually on the same side as the primary complex and is a hypersensitivity reaction. After this, the infection enters a latent stage which, in most people, lasts for the rest of their lives. In a minority, there is a fourth stage, which lasts about three years, until the primary complex is healed. This is the period of skeletal tuberculosis. Most cases appear during the first year, a large number during the second and the remainder during the third. In adolescents, this is also the period of pulmonary tuberculosis, most cases occurring within three years after the appearance of the primary infection. The age of the person is an important factor in determining the incidence of the conditions in the various stages.—*The timetable of tuberculosis, A. Wallgren, Tubercle, November, 1948, 29: 245.*—(*A. G. Cohen*)

Tuberculosis in Identical Twins.—Monozygotic twins, 22 years of age, had lived under identical conditions and had the same profession. One of the twins had a febrile episode during which roentgenograms revealed an infiltration in the apex of the right lung with a cavity in the right subclavicular area. Tubercle bacilli were found in the sputum. The other twin was examined as a contact and found to have an identical infiltrate with cavitation in the left lung; his sputum also contained tubercle bacilli. The parents had no roentgenographic evidence of chest disease but an older brother had had pulmonary tuberculosis treated by pneumothorax. After considering the problems of hereditary disposition to tuberculosis raised by these cases, the authors suggest that nurses with a family history of the disease be excluded from working on tuberculosis services. —*Sur les jumeaux tuberculeux, P. Braun & A. C. Maclouf, Rev. de la tuberc., 1947, 11: 707.*—(*V. Leites*)

Treatment of Tuberculous Glands.—The plan of treatment was to obtain healing and quiescence by medical means and then to remove the glands surgically. General measures included good food, rest, fresh air and ultraviolet light treatment. The patients usually remained ambulant. The head and neck were immobilized by means of a carapace. Fluctuant and cold abscess were in-

cised with a tenotomy knife; this usually did not lead to sinus formation. Enlarged tonsils were removed and attention was paid to the teeth. In addition, calciferol was given to 23 children and 11 adults. The first effect was a flareup which soon subsided. Of 14 children with draining sinuses, 3 were slow to respond; the others healed in one to three months. Of 6 adults with sinuses, 3 healed within three months and the other 3 more slowly. In every case, there was an increase in calcification and a definite decrease in periadenitis. The excised glands showed an acceleration of the normal healing processes. Before treatment, the red, white and differential blood counts, sedimentation rate, blood urea nitrogen and calcium and the renal function were determined. Toxicity of the drug was heralded by a rise in the blood urea, calcium and sedimentation rate. The daily dosage of calciferol for children was 100,000 units and for adults 150,000 units taken in tablet form. The total dosage reached 2 to 16 million units in children and 7 to 46 million in adults. Toxic symptoms of a mild degree were noted in 9 children and 2 adults, of a moderate degree in 2 children and one adult and severe in one adult. The earliest sign was constipation, followed by malaise, nausea, vomiting, anorexia, thirst and polyuria. There was no impairment of renal function in 11 patients and temporary impairment in 5. In one patient there was questionable permanent impairment.—*Calciferol in the treatment of tuberculous glands, S. Gauvain, Tubercle, November, 1948, 29: 259.—(A. G. Cohen)*

Diagnosis of Tuberculoderms.—(*1*) *Lupus vulgaris.* It occurs endemically in regions which have a moist, cool climate with relatively few hours of sunlight per year. It frequently begins in childhood and progresses very slowly. The first lesion is the so-called lupus nodule. It is a small, well circumscribed, red or brown infiltrate which cannot be obliterated by pressure. Later it has an apple jelly color, is covered by scales and may become fungoid in character. The histological picture is that of a tuberculous granuloma, occasionally with a central area of necrosis. *Lupus vulgaris* may appear on any part of the body but the face is an area of predilection. (*2*) *Lupus miliaris disseminatus.* This is a rare disease of sudden onset. The lesion consists of many discrete, noncoalescent, lupus-like nodules which appear within a period of six weeks. The nodules remain from three to six months and on healing leave a variola-like scar. At no time is there scale formation. Tubercle bacilli are demonstrated only with great difficulty; the tuberculin test may or not be positive. The eruption occurs almost exclusively on the face. (*3*) *Tuberculosis verrucosa cutis.* This is an inoculation tuberculosis usually encountered on the hands of physicians, veterinarians or butchers. The lesion is bluish red in color and forms scales early. The degree of verrucosity varies a great deal. There is always hyperkeratosis while caseation is absent. (*4*) *Tuberculids.* All tuberculids are probably hematoembolic processes. They are papular lesions, varying in size and secondary changes. The microscopic picture varies depending on the specific tissue response and the degree of secondary changes. The onset of this lesion is rather sudden. It usually has a self-limited course with a fairly rapid evolution.—*Criteria for the diagnosis of certain tuberculoderms, H. E. Michelson, J. A. M. A., November 6, 1948, 138: 721.—(H. Abeles)*

Gastric Tuberculosis.—Tuberculosis of the stomach is rare. Broders classifies the condition into (*1*) miliary tubercles of the stomach, (*2*) solitary tubercle, (*3*) tuberculous ulcer, and (*4*) pyloric stenosing tuberculosis. The author reports a case with miliary tubercles, ulcers and stenosis. The patient had epigastric pain, bloating, sour eructations and bouts of emesis. The stomach was resected and the patient died. At autopsy the lungs were free of tuberculosis. The stomach, duodenum and omental lymph nodes showed tuberculosis.—*Gastric tuberculosis, H. R. Morris, Am. J. Roentgenol., May, 1948, 59: 682.—(J. E. Farber)*

Tuberculosis of Stomach and Duodenum.—Two cases of tuberculosis of the stomach and one of the duodenum are reported. None of the patients had active pulmonary tuberculosis but abdominal lymphadenitis was a prominent feature. All 3 cases developed perforations or fistulous tracts at the site of their lesions. Roentgenologically there are no pathognomonic findings so that the ulcerative and infiltrative lesions can be confused with benign ulcer or carcinoma. The simultaneous involvement of the stomach and duodenum, the presence of fistulae or sinuses and signs of external pressure by enlarged lymph nodes are roentgenographic diagnostic findings suggesting tuberculosis even in the absence of any pulmonary lesion.—*Tuberculosis of the stomach and duodenum, H. W. Ostrum & W. Serber, Am. J. Roentgenol., September* 1948, *60: 315.*—(*J. E. Farber*)

Abscess after BCG Vaccination.—A 2 week old infant was vaccinated with BCG (0.04 mg.) in the right axilla by the subcutaneous method. He was transferred to another institution where his tuberculin test was found to be negative during the following three months. Revaccination was performed by scarification in the left axilla. One week later a small abscess developed at the site of the scarification and a few days later a similar abscess formed in the right axilla where the first dose of BCG had been given. Typical BCG cultures were obtained from both abscesses. The tuberculin reaction became positive shortly after the second vaccination. Thus, in this case bacilli had remained alive in the patient for three months without producing any clinical demonstrable local reaction and without rendering the tuberculin reaction positive.—*Petits abcès cutanés après revaccination par le BCG au siege d'une scarification récente et d'une injection sous-cutanée antérieure, J. Le Melletier, Rev. de la tuberc., 1948, 12: 235.*—(*V. Leites*)

Seedbed of Tubercle Bacillus.—The necropsy records of Bellevue Hospital patients over 15 years of age were examined for the period from 1935 to 1944. The following conclusions are made: Among patients who died from tuberculosis the clinical diagnosis was incorrect six times more often for persons over 50 than for persons under 30 years of age. Among patients who died from other diseases but who harbored communicable tuberculosis, there was clinical recognition of the tuberculosis in only one-fourth. Older persons can have progressive disease with so few manifestations of illness that they may not seek the advice of a physician and may remain unrecognized sources of infection. The registration of death from tuberculosis in New York City appears to be below the actual death rate with respect to persons over 50 years of age. In a high proportion of new cases of tuberculosis in adults in New York City, the disease is apparently contracted from unrecognized sources of infection. Mortality rates do not indicate either the extent or the location of the seedbed in which the tubercle bacillus resides. The seedbed may be greater in extent today than it was twenty years ago, for it is largely concentrated in persons, principally men, over 45 years of age and these are more numerous today than they were a generation ago.—*Disregarded seedbed of the tubercle bacillus, E. M. Medlar, D. M. Spain, & R. W. Holliday, Arch. Int. Med., April, 1948, 81: 501.*—(*G. C. Leiner*)

Pneumopericardium.—Two cases of pneumopericardium are described; both occurred in the course of therapeutic pneumothorax. One patient had had a pneumothorax on the left for five days; the other on the right for six and a half months prior to the appearance of the pneumopericardium. Neither patient had symptoms and the diagnosis was made by fluoroscopy. The pathogenesis remains obscure although a free pleuropericardial communication is suggested as an explanation. Puncture of the pericardium by the pneumothorax needle is unlikely. The presence of pneumopericardium was not considered a contraindication for the continuation of pneumothorax.—*Le pneumopéricarde,*

complication rare au course de la pratique du pneumothorax therapeutique, J. P. Nico, J. Lacorne & R. Comsault, Rev. de la tuberc., 1948, 12: 48.—(V. Leites)

Activity of Pulmonary Tuberculosis.—It is important to make a rapid and accurate estimate of the activity of minimal tuberculous lesions found in mass surveys. More than 75 per cent of these lesions have acquired stability at the time they are found. Activity can be determined accurately by roentgenologic examination and more rapidly and economically than by bacteriologic methods. In a series of 699 such cases followed for two to five years, activity was correctly determined from the first roentgenogram in 86 per cent of cases. A large film and an experienced roentgenologist are necessary. The photofluorogram should be used for screening only. Patients with minimal inactive pulmonary tuberculosis should be followed with roentgenograms at three month intervals for two years after discovery and at longer intervals thereafter.—*Accuracy of roentgen determination of activity of minimal pulmonary tuberculosis, C. C. Birkelo & P. O. Rague, Am. J. Roentgenol., September, 1948, 60: 303.—(J. E. Farber)*

Cavities "Below the Diaphragm."—The authors report 3 patients under pneumoperitoneum therapy whose pulmonary cavities appeared to be below the diaphragm. The cavities were located in lung tissue below the dome of the diaphragm in the anterior and posterior sulci, thus producing the illusion described.—*Pulmonary cavities "below the diaphragm," P. Morganstern & I. Pine, Am. J. Roentgenol., May, 1948, 59: 677.—(J. E. Farber)*

Surgical Collapse Therapy in Children.—This is a statistical evaluation of the results of treatment of tuberculosis by surgical collapse in children five to fifteen years old from 1932 to 1947 in Passy, France. Until recently this was the only French tuberculosis center where surgery was used extensively in children. Of 146 operations, 64 were phrenic interruptions, 44 thoracoplasties, 33 extrapleural pneumothoraces, 4 Monaldi drainages, and one was a cavernostomy. Over the fifteen year period the indications have gradually changed. In 1932, 68 per cent of the operations were phrenic nerve paralyses, 23 per cent thoracoplasties, and 9 per cent were extrapleural pneumothoraces. In the course of 1947, 78 per cent of the patients were treated with extrapleural pneumothorax, 15 per cent with phrenic paralysis, and 7 per cent with thoracoplasty. Extrapleural pneumothorax at present is considered the best method in children, 83 per cent of the patients having a "complete result." However, a great number of the operations are admittedly of too recent date to be evaluated as to long term results. No patient has been made worse by the intervention. There was one death several years later due to contralateral spread. Thirty-one thoracoplasties were performed after abandonment of an ineffectual pneumothorax; only 39 per cent gave "complete results." Thoracoplasty on 3 patients, seven to eleven years old, produced no deformity of the thorax in contrast to patients eleven to fifteen years old in whom deformity was very marked. The present indications for temporary phrenic nerve paralysis with or without pneumoperitoneum are limited to basal lesions of recent date and central location.—*La collapsothérapie chirurgicale chez l'enfant tuberculeux de cinq a quinze ans, H. Joly, Rev. de la tuberc., 1948, 12: 310.—(V. Leites)*

Coccidioidomycotic Pulmonary Cavitation.—Humans become infected with *Coccidioides immitis* usually by inhaling the chlamydospores and arthrospores of the fungus. Symptoms develop, after an incubation period of one to three weeks, in about 40 per cent of infected men. In the remainder the disease is completely asymptomatic. Women have a greater frequency of erythema nodosum and, therefore, a larger proportion have clinically manifest disease. The pneumonic or respiratory symptoms are of varying severity.

Pulmonary cavitation may develop in an area of coccidioidal pneumonitis or in a residual lesion some months after the primary infection. The patients with cavities rarely have disseminated infections and apparently possess effective immune mechanisms. The frequency of cavitation in asymptomatic coccidioidomycosis is not known; the incidence in hospitalized cases in the United States Army has been 2 to 8 per cent. The coccidioidal etiology of 274 pulmonary cavities was verified by recovery of the fungus in 40 per cent, by serological tests in 49 per cent, and by a positive coccidioidin without a tuberculin reaction in 11 per cent. Concurrent tuberculous and coccidioidal infections were present in 7 of the patients. The coccidioidomycosis was not progressive in any of these and the tuberculosis was progressive in only one. The relatively benign nature of the cavities is indicated by the fact that in the military patients three-fifths of them were discovered incidentally. The initial roentgenograms were made on almost three-fifths of the civilian patients because of hemoptysis which, however, was rarely severe. The other signs and symptoms of tuberculosis were rare. Ninety per cent of the cavities were single and 70 per cent were located in the upper portions of the lungs. In diagnosing these lesions, the coccidioidin skin test was the first step. Approximately 10 per cent of the patients reacted only to coccidioidin stronger than 1:100; a few did not react to 1:10. When the serological test was negative and a tuberculin reaction occurred, the diagnosis could be established only by recovering the fungus from the sputum or gastric contents. In three-fifths of the patients with fungi in the sputum, the etiology could also be demonstrated serologically. When detectable in patients with cavities, complement-fixing antibodies were usually present in low titer in contrast to the high titer characteristic of disseminated infection. A decline in the antibody titer and a decrease in the sedimentation rate were noted while cavities were forming. Many cavities closed quickly but some remained open for years. Bed rest may have aided closure early but was of limited value later. The risk of dissemination has been negligible and the possibility of contagion remote so that drastic intervention should be reserved for specific indications. Phrenic interruption has closed some old cavities. Pneumothorax should not be used for peripheral cavities because of the hazard of creating a bronchopleural fistula. Surgical removal of a persistent subpleural cavity by lobectomy or wedge resection safely eliminates the danger of spontaneous hydropneumothorax, which occurred in 2.6 per cent of the cases presented. Coccidioidal cavities and spontaneous hydropneumothorax are much less hazardous than the corresponding tuberculous lesions and incomparably less dangerous than disseminated coccidioidal granuloma.—*Pathogenesis of coccidioidomycosis with special reference to pulmonary cavitation, C. E. Smith, R. R. Beard & M. T. Saito. Ann. Int. Med., October, 1948, 29: 623.*—(*E. A. Rouff*)

Pneumonia with Salmonellosis.—An outbreak of food poisoning due to *Salmonella montevideo* was observed in 350 persons. Of these, 19 patients (6 per cent) developed bronchopulmonary manifestations. Four patients had acute bronchitis. One patient had bilateral bronchopneumonia; *Salmonella montevideo* was found in his sputum. Fourteen patients had clinical and roentgenographic evidence of interstitial pneumonia. All of them had an initial acute gastroenteric phase, lasting two to eight days. In 8 of the 14 cases there was a quiescent interval of one to five days, followed by increased toxicity, fever and bronchopulmonary complaints. In 6 cases there was no quiescent interval. The bronchopulmonary manifestations lasted from four days to two weeks. Sputum was cultured one to three times in 8 cases without recovering *Salmonella*. Sulfaguanidine, given during the gastrointestinal phase in 8 patients, and sufadiazine, given during the pneumonic phase in 3 patients, were without effect.—*Pneumonia associated with acute salmonellosis, A. P. Ingegno, J. B. D'Albora, J. N. Edson*

& P. J. Janquinto, Arch. Int. Med., April, 1948, 81: 476.—(G. C. Leiner)

Asbestosis.—Forty case of asbestosis, autopsied at the Medical College of South Carolina since 1930, are reviewed. The disease apparently did not progress long after exposure to asbestos dust ceased, although the fibrosis persisted and finally became scar tissue. In the milder cases there was little to indicate any influence of the disease on the health of the patient and death was usually caused by some unrelated condition. In practically all of such cases the asbestosis was undiagnosed until necropsy. This was also true in most of the cases of moderate lung damage. Asbestosis was conspicuous only when advanced and even then it was not always the principal or sole factor in the terminal illness. Asbestosis bodies remained in the lung indefinitely and were found twenty-seven years after the last exposure to asbestos dust. The location of these bodies in the terminal bronchiole and in the vestibular area of the lobule is significant in the pathogenesis of the disease. Bodies of smaller size appeared in the peribronchial lymph nodes, where there was foreign body reaction but usually little fibrosis. The presence of asbestosis bodies in the sputum indicated only that there had been inhalation of asbestos and did not characterize the condition of the lungs. The bodies may be found for years after the last exposure, but they may not be present in the sputum even in advanced cases of asbestosis. Pleural involvement was not constant and was probably a secondary occurrence. Local, fibrotic, nodular lesions similar to those characteristic of silicosis occurred frequently, which suggests that they may have been produced also by the asbestos. Carcinoma of the lung was found in 7.5 per cent of the abestosis cases as compared with a general incidence of one per cent, suggesting that the two conditions are related. Tuberculosis of the lung was found more frequently than in general autopsy series but careful analysis fails to show any direct relationship.—*Asbestosis: Analysis of forty necropsied cases, K. M. Lynch & W. M. Connor, Dis. of Chest, November-December, 1948, 14.—(E. A. Rouff)*

Heart in Bronchial Asthma.—The cardiac and circulatory changes were studied in 301 patients with bronchial asthma. Physical examination revealed various degrees of emphysema in 61 per cent. Roentgenograms confirmed the frequency of so-called "drop-heart" with emphysema. It was present in 14 per cent of the patients without any auricular or ventricular enlargement. Although electrocardiography was often valuable, it was difficult to detect slight changes in the heart by this method. The most conspicuous electrocardiographic changes were (*1*) right axis deviation, (*2*) peaking and widening of the P waves in leads II and III, (*3*) depression of the ST segment with inversion of the T waves in leads II and III, (*4*) splitting of the R and S waves, and (*5*) prolongation of the QRS conduction time. Right axis deviation was present in 44 per cent of the cases, left axis deviation in 28 per cent, and no axis deviation in 28 per cent. Uncomplicated bronchial asthma did not change the cardiac configuration. Associated cardiac disease, pulmonary congestion, or structural deformity were responsible for enlargement of the pulmonary conus and arteries, as well as for right cardiac hypertrophy with electrocardiographic axis deviation. Any "aortic or mitral configuration" was a sequel to valvular heart disease and not the result of bronchial asthma.—*The heart in bronchial asthma, M. K. Hajos, Ann. Allergy, November-December, 1948, 6: 655.—(B. Hyde)*

Commercial Nebulizers.—The Council on Physical Medicine of the American Medical Association has accepted only 3 nebulizers now commercially available, the de Vilbiss Nos. 40 and 44 and the Holmspray No. 630. Physicians have recommended other designs but there is little information available about their characteristics. This study compares the performance of several nebulizers now being sold. Failure of the apparatus to per-

form satisfactorily was due to (*1*) the production of an insufficient amount of mist, (*2*) too many large particles which did not get beyond the mouth and pharynx, or (*3*) a preponderance of true vapor or of particles so small that they were exhaled. Nebulizers containing small amounts of distilled water were weighed before and after 500 compressions of the hand bulb, and the weight delivered by one compression was calculated. This weight varied in the different nebulizers from 13.0 to 0.4 mg. The "Breathing" nebulizer delivered the most mist followed in the order of decreasing delivery by "Asthma Neprin" and "Vaponefrin," "De Vilbiss No. 40," "Vapto," "Broemmel," "Peralta," "Felrodo," "Parke-Davis Nebulizer," "De Vilbiss No. 44," "Defender," "Stearns," "Parke-Davis Adrenalin Vaporizer." The "Breatheasy" discharged large droplets which lodged on the posterior pharyngeal wall. "Vapto" produced particles of a size reaching the upper respiratory tree and "Broemmel", a fine mist. "Parke-Davis Nebulizer," "Defender," and "Stearns" emitted droplets.—*A comparative study of commercial nebulizers, G. F. Harsh, Ann. Allergy, September-October, 1948, 6: 534.*—(*B. Hyde*)

"Nechaphyl" for Bronchial Asthma.—Thirty-five of 40 patients with bronchial asthma obtained relief from "nechaphyl with phenobarbital" which they judged to be at least as satisfactory as that obtained with various oral ephedrine preparations. No toxic manifestations were noted and "nervousness" was diminished. "Nechaphyl" alone was less satisfactory than oral ephedrine.—*Nechaphyl in bronchial asthma, S. W. Simon, Ann. Allergy, November-December, 1948, 6: 662.*—(*B. Hyde*)

Apical Lung Cancer.—Primary carcinoma of the lung located in the apex (Pancoast tumor) produces a characteristic clinical and roentgenographic picture. Clinically, the patient complains of shoulder girdle pain which later radiates down the inner side of the arm into the fourth and fifth fingers; the affected shoulder droops. The upper eyelid on the side of the lesion becomes ptotic and perspiration is absent over that side of the face and head (Horner's syndrome). Cough is generally absent. A mass can be palpated in the related supraclavicular area. Over the apex there is percussion dullness and diminished breath sounds. Roentgenograms reveal a well-defined homogeneous shadow of increased density high in the apex. The trachea is displaced and slightly indented. These tumors should be completely removed before extension to the mediastinum or brachial plexus has occurred. Radiation therapy has been disappointing but should be prescribed for all inoperable cases.—*Primary cancer of the lung with special reference to apical lung tumors, J. J. Stein, Am. J. Roentgenol., July, 1948, 60: 58.*—(*J. E. Farber*)

Mediastinal Cystic Hygroma.—Cervical hygromas are not rare. On the other hand, the literature contains reports of only 19 cases of cervicomediastinal and 8 of intrathoracic hygromas. To these, the authors add 3 cases of their own. The condition is a congenital malformation arising during the development of the lymphatics by budding. The mechanism whereby it appears in the chest is uncertain. Pathologically it is a thin-walled cystic structure lined by endothelium, and is usually multilocular. The fluid in it is thin. A variety of structures such as connective tissue strands, fat, blood vessels and nerves may be found within it. Clinically, hygromas of the neck are usually noted at birth; they do not cause symptoms unless they displace the trachea or esophagus. They are thin-walled and translucent. In the cervicomediastinal type, roentgenography shows a shadow in the upper chest on the side of the mass; this varies in size with crying and other activities. The mediastinal type is found only on routine roentgenologic examination and is difficult to diagnose precisely. Surgical excision is indicated for cosmetic reasons and to avoid infection or tracheal compression. The cervical portion is removed first. The intrathoracic portion is then sclerosed with

chemicals. — *Cervicomediastinal and mediastinal cystic hygromas, R. E. Gross & E. Hurwitt, Surg., Gynec. & Obst., November, 1948, 87: 599.—(A. G. Cohen)*

Traumatic Hemothorax.—World War II taught that prompt and repeated aspiration of traumatic hemothrax is the immediate treatment of choice. The purpose of this is to obviate the formation of adhesive fibrinous pleuritis. The latter may result in limitation of lung function or failure of re-expansion. Six exemplary cases are reported.—*Important sequelae and complications of hemothorax resulting from penetrating wounds of the pleural cavity, S. P. Barden, A. J. Roentgenol., April, 1948, 59: 525.—(J. E. Farber)*

Intrathoracic Injury.—Five per cent of 4,000 battle casualties admitted to one army hospital had wounds involving the intrathoracic structures. Roentgenograms should reveal (*1*) the presence and location of foreign bodies, (*2*) the extent of bony injuries to the ribs, spine and sternum, (*3*) lesions of the lungs, (*4*) lesions of mediastinal structures, and (*5*) the status of the pleura and the contents of the pleural space. Foreign bodies are frequently seen in the lung with little or no evidence of reaction in the lung or pleura. Many are arrested in the hilar and mediastinal structures. Traumatic lung lesions are usually small, circumscribed, and resolve uneventfully. Air and blood in the pleura are absorbed rapidly especially if promptly aspirated. Aspiration prevents thickened pleura and pleural infection. Mediastinal foreign bodies may escape detection and require overpenetrated films. Pericardial effusion suggests pericardial trauma.—*Roentgenological manifestations of intrathoracic injury due to missiles, W. A. Evans, Jr., Am. J. Roentgenol., May, 1948, 59: 662.—(J. E. Farber)*

Roentgenography of Hydatic Disease.—Closed hydatid cysts of the lungs differ roentgenographically from the open ones. The closed cyst, which usually does not contain daughter cysts, appears roengenographically as a round homogeneous shadow in the lung parenchyma. The intercostal spaces may bulge. As the cyst grows, it often loses its roundness and becomes polycyclic, being more often visible in lateral views. The sharp limits disappear due to a thickening of the pericystic membrane. Echinococcus causes displacement of the bronchi that surround the cyst but so do all benign tumors; bronchography is thus of little diagnostic help. Flurosocopy is also of little help, but the existence of large round shadows in both lungs is diagnostic of hydatid cyst. If a cyst bursts and the level of the liquid is visible, the roentgenographic diagnosis is assured. Pericystic emphysema after rupture of a cyst signifies death of the parasite. Clinically, this phase manifests itself by small hemoptyses. The sign of the double arch occurs only rarely after rupture of a cyst and is a hydropneumocyst with floating membrane. At times rupture of a cyst causes widespread inflammation and atelectasis of large areas of the lung. Some of these pneumocysts (or hydropneumocysts) disappear without treatment; others disappear following treatment including drugs, pneumothorax or surgery.—*Hydatid disease and its roentgen picture, P. M. Schlanger & H. Schlanger, Am, J. Roentgenol., September, 1948, 60: 331.—(J. E. Farber)*

DIAGNOSIS AND TREATMENT OF LUNG TUMOR[1,2]

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INTRODUCTION

Carcinoma of the lung accounts for between 6 and 8 per cent of all cancer and approximates the frequency of gastric carcinoma. It is, therefore, a common disease which every general physician may expect to encounter in his practice. Usually the diagnosis is easily made by observation and analysis of the symptomatology which differs little from that of numerous other thoracic diseases.

The results of treatment are comparable to those of other visceral cancers and much progress has been made in treatment in the past fifteen years, both in respect to earlier diagnosis and improved technique. There has been a marked reduction in the immediate operative mortality, which formerly was very high but now is less than 5 per cent. The chief problem, in the present state of knowledge or lack of knowledge of cancer causation is that common to all malignant disease, i.e., earlier diagnosis. The available surgical procedures are now about as radical as is feasible or practically applicable. Some improvement still is possible in short term results on poor risk patients by further refinement in methods of preparation and of after care.

The content of this paper is limited to a statement of working policies in the detection of cases of lung tumor and the care of patients with such disease; in other words, practical diagnosis and treatment based on my own observations and experience. It excludes detailed statistical reporting of factual data as garnered from records of some 385 microscopically proved cases. It also excludes minutiae of technical detail as both these items are of especial concern only to those primarily interested in this single disease.

MEASURES TO MINIMIZE INCIDENCE

This topic is mentioned only because there is nothing definite to report, nor is there likely to be until the cause of cancer is discovered. Cigarette smoking has been incriminated, but not proved. Neither has chronic inflammation or irritation been shown to be causative, as is suspected in relation to carcinoma of the cervix. The one exception to complete ignorance of cause is the established fact of increased incidence in miners exposed to radioactive ores. The reason for the 4 to 1 male predisposition also is not explained.

DIAGNOSIS

Value of Population Survey

Large scale radiographic screening has revealed some type of thoracic lesion in approximately 1 per cent of the segments of population examined, but only

[1] From the Good Samaritan Hospital, Woodbury, Tennessee.

[2] Presented at the Southern Tuberculosis Conference, Savannah, Georgia, October 1, 1948.

a fraction of these cases have been shown or even suspected of having carcinoma. The group in which carcinoma is most prevalent is not often reached by the screening policies adopted for such surveys, which are designed to find tuberculosis and are directed toward the younger age groups. However, an occasional case of carcinoma is found.

Furthermore, periodic health examination will not find the silent malignant lesion in the lung, unless: (*1*) it is accompanied by roentgenologic examination and the growth is in the lung parenchyma or encroaching on the bronchial lumen; and (*2*) such examination is repeated more often than is necessary in Public Health control programs. The three most valuable approaches to discovery of really early carcinoma of the lung are: (*1*) education of the public to see a physician for any cough of over two weeks' duration; (*2*) education of the profession to inquire for symptoms and at least fluoroscope every new patient and make chest films on fluoroscopic indication or presence of symptoms; (*3*) stop the practice of symptomatic treatment of cough or pleurisy without evaluation of cause.

Symptomatology

In contrast to carcinoma of the stomach, lung cancer usually causes early symptoms, but they are often overlooked. They are so ordinary in character that both the patient and the physician are likely to disregard them in early stages. As determined from study of one group of 157 consecutive, microscopically proved cases (1), the early symptoms in the order of frequency are:

Cough: Cough is present in 93 per cent of the patients with lung cancer. In the early states it is dry or irritative, but soon becomes productive of mucoid or purulent sputum. Most often it is attributed to cigarette smoking or a mild cold.

Pain: Almost every patient has a different concept of pain, depending upon his temperament and racial and social background. Early carcinoma of the lung rarely causes true pain, but some degree of pleuritic or vague thoracic discomfort will be admitted in slightly over half the patients.

Sputum: The character of sputum in carcinoma of the lung is related closely to secondary infection. Very early in the disease true mucoid sputum, apparently caused by mucosal irritation or stimulation by the growth, is encountered. Later the sputum usually becomes purulent, but not foul or fetid, unless there be bronchial obstruction or tumor necrosis.

Hemoptysis: Blood spitting which occurs each day, every day, persistently over a period of time, is caused almost solely by carcinoma. However, hemoptysis in carcinoma also may be sporadic or intermittent.

Wheeze: This symptom or sign is not to be confused with asthma. It is an indication of bronchial encroachment to a point of incomplete occlusion and its intermittent character in early stages is explained by the presence or absence of secretion in the narrowed bronchial lumen. In combination with one or more of the preceding symptoms, it provides strong presumptive evidence of bronchial occlusion caused by tumor unless there be a history of foreign body inhalation.

Confirmation of localized obstruction may be furnished by roentgenologic observation of mediastinal shift or localized compensatory emphysema.

A late symptom of *dyspnea* is closely correlated to *pleural effusion.* Bloody effusion is the type commonly found, but is not the infallible indication of malignancy once thought, because it can also be caused by infarct or an inflammatory reaction. Furthermore, effusion also occurs in nonsanguinous form from pleural involvement by cancer.

Severe pain, as distinguished from mild pleural discomfort, should be regarded as a consequence of brachial plexus, chest wall, or mediastinal invasion. Relief from such pain is infrequently obtained by direct operative procedures, including removal of ribs, intercostal neurectomy, or lobar excision. Occasionally, X-ray therapy is of transient benefit as are the nitrogen mustards. On the whole, if the pain really remains intractable and beyond the range of control by medication, one must resort to chordotomy or frontal lobotomy. The latter procedure has given very gratifying results to a few of our cases but, because of the attendant personality changes, must be embarked upon only after full consultation with the neurosurgeon, the personal physician, and the family.

Hoarseness: The appearance of hoarseness or huskiness of voice suggests paralysis of the recurrent laryngeal nerve from mediastinal extension of the growth. Occasionally the voice will return almost to normal after a few weeks of hoarseness, giving the patient and perhaps the physician a false sense of security, even though the vocal cord remains paralyzed. This phenomenon, when it occurs, is related to laryngeal compensation. In most instances the voice remains permanently husky. One must beware of deciding forthwith that a patient has inoperable cancer of the lung when he has been shown to have an abnormal shadow in the chest and a paralyzed vocal cord. We have now encountered 3 cases with this situation in which cancer was not present and the patients made a complete recovery following appropriate surgery. One was a dermoid cyst with infection; one was a teratoma with surrounding inflammatory reaction; and one was a substernal goiter into which there had been recent hemorrhage.

Weight loss and weakness occur in later stages of the disease and direct attention to a careful search for peripheral nodes and enlarged liver or other signs of distant metastasis.

Physical Signs

The physical signs of lung cancer are related solely to bronchial obstruction and sequelae or to extension of the growth. The presence of a tumor mass itself is rarely detectable by physical examination. One may find the signs of pulmonary infection or atelectasis or pleural effusion. Paralysis of the phrenic nerve or the recurrent laryngeal nerve are detectable by physical observation, as previously mentioned. Every case should have careful, methodical palpation of the supraclavicular fossae and axillae for nodes. In instances where pain is a complaint, it is important to palpate meticulously the intercostal spaces and muscles about the scapula for signs of tenderness or thickening from tumor invasion.

Definitive Diagnostic Tests

The observations thus far mentioned are limited to recognition that disease is present, but are not definitive, for they may be encountered in various types of disease of the lung. Further study should include as many of the following additional procedures as are required for diagnosis.

1. Examination of the vocal cords with a laryngeal mirror. This is not a specialists' procedure and the simple technique can be mastered by anyone with a little practice.

2. Fluoroscopy. This should be a part of every general examination, and from it one gains specific information concerning diaphragmatic movement and type of movement. On fluoroscopy it is possible to detect immediately paradoxical movement or high stationary position of the diaphragm, secondary to a paralyzed phrenic nerve from tumor extension into the anterior mediastinum. The phrenic nerve, however, can be paralyzed by inflammatory reaction or by the pressure of benign lesions such as an aneurysm of the aortic arch. One observes the position of the mediastinum; whether there is shifting with breathing; whether the shift is toward or away from the lesion; and whether the position of the mediastinum is variable or fixed. Localized emphysema is sought and the respiratory phase in which it appears is noted. Partial bronchial obstruction causes localized expiratory wheeze and emphysema in combination. From fluoroscopy one may better determine what special radiographic views will best record the lesion. Finally, one learns whether an abnormal mass is of fixed size, pulsatile, or expansile.

Radiographic films are obligatory in the presence of symptoms or signs even if fluoroscopic observations are normal because the problem at this stage is one of differential diagnosis. Fluoroscopy alone may fail to reveal the early infiltrative, or exudative, or very small lesion, because of limited optical resolving power. It is beyond my competence to speak authoritatively on the purely technical phases of radiology, but the following common errors in making radiographs are mentioned because they are responsible for some errors in diagnosis: (*1*) omission of the lateral films which often assist in detection of pathology behind the heart or near the mediastinum and permit positional localization to a pulmonary segment, division or lobe; (*2*) failure to obtain oblique views of lesions requiring differentiation from anomalies or morbid processes in the heart or great vessels; (*3*) simple, technical inadequacy, such as overexposure and underdevelopment, gross underexposure or development in solutions that are too hot.

Supplementary films with heavy exposure or with the Buckey diaphragm should be made for lesions of marked or extensive density in order to detect an internal cavity or to visualize the bronchial lumen, and to study the ribs, pleura and mediastinum for irregularity. Laminograms occasionally are helpful but are more useful in the study of tuberculosis.

Radiographic Evidence

The most direct primary evidence consists of the demonstration of a tumor mass. If circular in form, it is likely to lie in the periphery, even though it may

appear to be situated close to the hilus on a postero-anterior film. A lateral film will complete definition of the location of the mass. If multiple shadows of round masses are seen, metastatic disease to the lungs is naturally suspected. If the mass lies in the periphery, is round, and is more than 4 cm. in diameter, either adenocarcinoma or solitary metastasis from a hypernephroma is to be suspected, and intravenous pyelograms should be done. The other primary evidence consists of the presence of a blocked bronchus with an ovoid shadow of tumor projecting into the bronchial air column above the obstruction. This is sometimes seen on a plain film, more often on a Buckey film, and most often on a laminograph.

The secondary evidence of the presence of a tumor usually consists of radiographic and the other signs of pulmonary atelectasis or infection which are related to variable degrees of bronchial encroachment and obstruction. Important information, which suggests the occurrence of subcarinal lymph node extension, is afforded by a broadened or distorted tracheal bifurcation.

Certain axioms are of aid in radiographic interpretation, remembering that they are statements of clinical experience and not of mathematical law. Localized emphysema and wheeze in an adult are usually caused by a tumor. A widened carina indicates disease in the mediastinal nodes and hence is extremely likely to indicate the presence of tumor. A lung abscess with thick wall (greater than 1 cm.) and nonfetid sputum is presumptive evidence of carcinoma. A fluid "level" with upward convexity is caused by central necrosis of a tumor mass having bronchial communication. Radiographic reports reading "suggest recheck in two months to rule out carcinoma" are never justified on the basis of haphazard or incomplete study, and may cause needless delay in diagnosis.

Summary of radiographic study: Radiographic examination excels all other methods in the detection and recognition of lung cancer if done completely and properly by persons of experience and competence.

Examination of the Sputum for Cancer Cells

Examination of the sputum for cancer cells is an old method which has been in use for many years. It has recently been repopularized and increased in value by the work and writing of Papanicolaou. Such examinations should be performed in all cases under suspicion of carcinoma of the lung, just as a search for tubercle bacilli in the sputum is made in all cases under suspicion of pulmonary tuberculosis. Experienced observers make very few false positive reports, but the report must be correlated with other observations. Negative reports or findings have little significance. The test is of considerable value when positive, but is not as yet as definitive as the evidence provided by examination of material obtained by a direct biopsy.

Thoracentesis is indicated in cases in which a pleural effusion is detected on clinical or radiographic examination. Aspiration is best done with a short beveled needle of number 18 gauge lumen, 2 inches in length. One might easily recite a series of cases seen with complications of pleural or pulmonary infection, hemorrhage or spread of disease, which were direct sequels to the injudicious use of a large, long, bayonet-pointed needle.

The fluid obtained from undiagnosed cases should be studied by smear; by appropriate cultures for aerobic and anaerobic microorganisms, including the fungi; determination of specific gravity and cell count; concentration and examination for tubercle bacilli and cancer cells. In cases of known carcinoma, when the question is reduced to one of malignant pleuritis, study of the concentrate by cytologic methods alone is sufficient. Although usually true, the long held tenet that the presence of pleural effusion with carcinoma invariably indicates inoperability has had to be altered. Similar effusions may be caused by associated or secondary parenchymal infections. A three year respite from symptoms has been observed after resection of a pulmonary carcinoma in which there was a massive pleural effusion.

On rare occasion the adjuvant use of a thoracoscope is helpful. Likewise, replacement of fluid with a mantle of air now and again will assist in demonstration of pleural nodularity on radiographic examination.

While considering the question of the passage of instruments through the chest wall, punch biopsy deserves mention. This procedure was formerly more frequently used by us than at present. It may be accompanied by many hazards such as hemorrhage, infection, or spread of carcinoma along the needle tract. Moreover, the findings are frequently negative when carcinoma is known to be present. Punch biopsy should be used only in cases deemed inoperable but on which a microscopic diagnosis is necessary as a guide in nonsurgical treatment.

Bronchoscopy

Next to roentgenologic study, bronchoscopy is the most important definitive procedure in evaluation of the patient with possible lung cancer. Evaluation is used as a broader term than diagnosis. If the bronchoscope is employed solely as a lighted tube through which someone may pass a forceps and excise a piece of abnormal tissue if fortuitously encountered, the value of the endoscopic study is limited in the same fashion as if the roentgenologic study were considered to be completed by the taking of a single postero-anterior chest film.

Many reasons exist for omitting bronchoscopy, including a peripheral location of the lesion, the absence of bronchial obstruction, or the ability to establish a diagnosis and decide upon therapy by other means. The indications for performance of bronchoscopy are symptoms, signs or roentgenologic findings of bronchial encroachment or obstruction.

The endoscopist must observe the mobility of the vocal cords and any variation from the normal in the size, contour, shape, flexibility, direction and position of the trachea, carina, primary lobar and segmental bronchi. He must note the appearance of the bronchial mucosa and the location and amount and type of secretion present. Furthermore, he must observe and record the exact position and gross appearance of any tumor found, with particular thought to the bearing of such information on operability. The endoscopist takes a biopsy of suspicious tissue. He examines the upper lobe bronchi, on indication, with optical systems that permit retrograde or right angle visualization. In addition, he may irrigate and aspirate secretions by use of appropriately curved flexible instruments from any segmental bronchus in the lung. Such fresh secretions immediately

prepared in the laboratory may yield well preserved cells from which the pathologist can accurately determine a diagnosis. Negative evidence from endoscopic examination so conducted is of equal importance.

So handled by a seasoned endoscopist, acting as a consultant with clinical acquaintance with the case, one may anticipate on the basis of our experience the following results in approximate figures. Eighty per cent of eventually proved cases of lung cancer will be bronchoscoped. Ninety per cent of the cases bronchoscoped will have the diagnosis established thereby or stated as a positive suspicion from the collateral observations which the bronchoscopist makes. If bronchoscopy is handled as a purely laboratory procedure, to be done by a technician with no knowledge of the case, under remote "pushbutton-like" control from a clinician, the results, I should think, would be less rewarding.

Bronchography rarely should be done in the diagnostic study of lung tumor. The oil may be trapped behind a tumor with production or accentuation of pneumonitis. It carries a slight risk of causing spread of tuberculosis. Moreover, bronchography confuses the radiographic appearance of early inflammatory disease and may make further accurate observation of progress either tedious or inaccurate. Bronchography is a technique for outlining the pattern of disease in bronchiectasis, and even there is only incidentally of diagnostic value.

Thoracotomy

An occasional case is enountered in which no diagnostic approach other than thoracotomy will be decisive. We have formerly made the statement that one rarely learns more about the diagnosis on opening the thorax than was known beforehand, although of course the extent of disease can be determined with finality only through the open thorax. This attitude was a reasonable one when the risk of operative mortality was high. Two facts have caused alteration of this viewpoint. First, the risk of thoracotomy itself now approaches zero and, secondly, more pulmonary lesions are being discovered in an asymptomatic stage. Therefore, thoracotomy and excision of the lesion has become preferable to assuming the risk that a mass of unknown nature is carcinoma. At thoracotomy one does not cut into the lesion, but excises it; the "biopsy" sometimes even becomes a lobectomy. For processes not subject to diagnosis by other methods, such a course is believed wise and conservative. It is excellent treatment for the fibrocaseous tuberculous mass before cavitation has occurred. It offers the best chance for cure of really early carcinoma. It does not unnecessarily sacrifice lung tissue. In certain instances it relieves general worry about the diagnosis and the patient's future. What happier outcome could be asked than the removal of a treacherous appearing pulmonary lesion, with minimal risk of complication or fataliy, and the finding that it is a benign lesion about which there need be no further concern?

Common Sense

Thus far in the discourse I have emphasized scientific exactitude in diagnosis and discussed the various methods for its attainment. I now wish to stress common sense, not as a substitute for, but as a corollary to, the other measures.

It is admittedly desirable to pursue every tumor problem to the attainment of a morphologic answer, and toward such a conclusion at one time I unremittingly worked. With the aging process, however, has come better realization of certain facts of life. One of these is that a balance must be struck between the value to the patient of a given exact diagnosis and the benefit the patient is likely to receive from the mere fact of proof. Otherwise, patients with advanced and inoperable cancer may be submitted to tedious and expensive diagnostic procedures having little justification beyond providing laboratory proof of what is already clinically apparent, since hopeful therapy is not expected to be in any way altered thereby.

A second realization is that the so-called "only hope" type of operation, done with forethought that the cancer is advanced and beyond surgical eradication, usually proves a snare and a delusion for the patient, the surgeon, the family, and the personal physician. I wish not to be misunderstood. I favor and practice a radical and aggressive attitude in the treatment of carcinoma by surgery; but one wishes to avoid a foolish attitude. There is often strong psychological or social pressure on the family, transmitted to the surgeon directly or through the family physician, to "do everything possible," which really means "do everything helpful." Every honest surgeon naturally wishes to do this but must assume responsibility for deciding (often a lonely decision) whether the operation may be helpful or merely possible.

A third realization is that the palliative operation must really earn its name, in respite from suffering, to merit much use. Undoubtedly, this procedure has a place in the treatment of certain cases with a sequestrating lobe, copious foul sputum, and hemorrhage. But fine discrimination is required to select those whose remaining weeks or months may be made easier. To others, one may have but added to an already sad situation the burden of an operation, with its attendant pain and expensive hospitalization.

In summary of this heading entitled "Common Sense," I urge that greater diagnostic effort, intensified education of the laity, and more surgical aggressiveness be directed toward early, silent or incidentally discovered pulmonary lesions; and less of these efforts toward the late, advanced lesions.

TREATMENT

X-ray Therapy

Selectively applied X-ray therapy makes a few patients more comfortable. Widely or indiscriminately used, the average benefit fails to justify the discomfort, inconvenience, and expense of application. Its usefulness is greatest in undifferentiated, high grade epidermoid, and oat cell types. In these specified types, surgery does not alter the outlook in any demonstrable way. Pleural effusion quite often responds dramatically to X-ray therapy. An occasional patient will experience relief from severe pain. In these, the relief usually is apparent before excessive therapy has been given.

Radium offers no advantages over X-ray therapy and the facility with which

it can be applied causes X-ray therapy to be chosen in most cases to be treated by actinic radiation.

Palliative Resection

Palliative resection has been already discussed in part. It is recommended only for cases burdened with complications of secondary infection. The duration of life until death from carcinoma is in no way changed, but the patient may be prevented from dying of infection or hemorrhage and has a more comfortable terminal period.

Surgery for Eradication of Disease

At the present time complete surgical extirpation offers the only hope for cure. Pulmonary resection, including both pneumonectomy and lobectomy, in our hands now carries a 5 per cent hospital mortality risk on the basis of a 30 per cent resectability rate (1). It is my belief that lobectomy for selected upper lobe lesions and peripherally placed lower lobe lesions, particularly in older patients, is as satisfactory a procedure as pneumonectomy. Moreover, lobectomy is associated with a lower operative hazard and better subsequent functional status. For the majority of cases, however, a radical pneumonectomy is preferable.

A discussion of technical details is not considered germane to this presentation. It is assumed that the surgeon and anesthetist in charge of a given case are competent to handle such matters. It is my understanding that this audience, in respect to surgery, is more interested in what surgery should be done and why it should be done, than in exactly how it is done.

Results of Surgical Treatment

No one, however, is more interested than you in what surgery has or can accomplish. The hospital mortality from pulmonary resection for carcinoma has now been reduced to 5 per cent. Our resectability rate has been 31.8 per cent. Resected cases have a 64 per cent chance of being alive and well one year later. Rarely does an untreated case live a year after diagnosis. It was previously reported (1) that 23 per cent of the patients subjected to pulmonary resection more than five years previously were found to be living and well at the end of five years. The predominant factor affecting survival after resection was there shown to be the microscopic cell type. In this series, epidermoid carcinoma had the least gloomy prognosis, followed in sequence by adenocarcinoma, oat cell and undifferentiated types. In fact, no patient with an oat cell or undifferentiated carcinoma lived more than a year after resection. Duration of symptoms, the size of the lesion, the type of resection done and the presence or absence of involvement of lymph nodes were found to exert significant but not determinative effects upon the survival time.

OTHER LUNG TUMORS

Aside from adenoma, other varieties of lung tumor are rarities. I know of two examples of primary sarcoma and have personally treated one case of myxo-

chondroma. Unless the rare hamartoma is to be so classified, I know of no other types of lung tumor. Adenoma occurs with approximately one-tenth the frequency of pulmonary carcinoma. At least 80 per cent of the cases are found in women, and particularly young women. The rate of growth of adenoma is slow. Cough and intermittent, sporadic hemoptyses are the common symptoms and these may often be controlled for years in a particular patient by periodic bronchoscopic removal of the intralumenal portion of the tumor. The tumor, however, has a striking propensity for extralumenal growth which attains a size grossly disproportionate to the intralumenal projection. As a consequence, the gradual development of bronchiectasis is a common sequence that eventually requires lobectomy. Over 90 per cent of patients with adenoma are alive and well for five and more years subsequent to treatment. In fact, the only mortality of which I am aware among 24 cases is that of one patient who died immediately postoperatively.

Regardless of who is right in the academic argument which has proceeded for several years on the question of whether bronchial adenoma is a malignant tumor or not, one may again find comfort and security in a common sense viewpoint. The expectancy for a patient who has an adenoma removed is that she will be well and remain well thereafter. The finding of, or the failure to find, cells resembling adenoma cells in adjacent lymph nodes has, in my experience, had little, if any, bearing on prognosis. From the standpoint of the clinical course of illness, the fact is that adenoma is a different tumor entity from carcinoma and survival rate statistics concerning the two disease must be scrupulously segregated if accuracy of reporting is to be maintained.

SUMMARY

Cancer of the lung accounts for about 7 per cent of all cancers. Results of treatment are comparable to other visceral cancers. The hospital mortality rate for surgical resection is now under 5 per cent. Diagnostic methods are reviewed and evaluated. Mature judgment seasoned by common sense should be applied in the avoidance of burdensome and nonbeneficial surgery for advanced, hopeless cases. Complete surgical extirpation offers the only hope of cure at the present time.

Twenty-three per cent of cases resected more than five years were found to be living and well at the end of five years. Bronchial adenoma occurs with one-tenth the frequency of lung cancer but has at least nineteen times the survival expectancy. Clinically, it is a different entity from carcinoma, whatever its pathologic classification according to degree of malignancy may eventually prove to be.

SUMARIO

Diagnóstico y Tratamiento de los Tumores Pulmonares

Al cáncer pulmonar corresponden aproximadamente 7 por ciento de todos los cánceres. Los resultados del tratamiento se comparan a los obtenidos en otros cánceres viscerales. La mortalidad hospitalaria para la resección quirúrgica es

hoy día inferior a 5 por ciento. Después de repasar y justipreciar los métodos de diagnóstico, señálase que hay que aplicar el juicio maduro templado por el sentido común a fin de evitar la cirugía molesta y estéril en los casos avanzados desahuciados. Las únicas esperanzas de curación se cifran actualmente en la extirpación cruenta total.

Veintitrés por ciento de los casos resecados hace más de cinco años se hallaban vivos y sanos al cabo de cinco años. El adenoma bronquial no alcanza en frecuencia más que la décima parte del cáncer pulmonar, pero muestra una expectativa de sobrevivencias a lo menos diecinueve veces mayor. Clínicamente, representa una entidad distinta del carcinoma, no importa la clasificación patológica que reciba con el tiempo, conforme al grado de malignidad.

REFERENCE

(1) Adams, R.: Primary lung tumors, J. A. M. A., March 2, 1946, *130*, 547.

OBSERVATIONS ON THE CAUSES OF DYSPNEA IN CHRONIC PULMONARY GRANULOMATOSIS IN BERYLLIUM WORKERS[1]

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INTRODUCTION

Chronic pulmonary granulomatosis in beryllium workers has been characterized by the delayed onset of weight loss, with or without anorexia, irritative coughing, exertional dyspnea, intermittent chills and fever following a known or suspected exposure by inhalation to beryllium compounds in a finely divided state (1 to 6). The most outstanding symptom has been exertional dyspnea, and there is often marked loss of weight. Clinical examination of these patients usually reveals variable degrees of respiratory distress even at rest, consisting of tachypnea, dyspnea, orthopnea and coughing. Cyanosis and clubbing of the nails may be present. Chest expansion is often limited and the vital capacity reduced. Signs of moisture may be detected by auscultation of the lungs, and the chest roentgenograms characteristically show diffuse bilateral miliary nodular shadows and reticular markings in the lung fields. Prominence of the right auricle and the pulmonary conus is noted in extreme cases. The course is one of delayed recovery in a few (7), gradual progression in many, and eventual death from right heart failure in almost a third of the patients (1, 2, 5). There is no alteration of the course by any known therapeutic agent, but supportive oxygen therapy appears to prolong life.

Preliminary respiratory studies (8) have shown that overbreathing during activity due to loss of breathing reserve and impaired pulmonary diffusion of oxygen are the earliest evidence of lesions. Arterial hypoxemia, secondary to an increased alveolar-arterial oxygen gradient, is inadequately compensated by hyperventilation especially during exertion. Advanced cases also show reduction in maximum breathing capacity together with emphysematous changes. Pathological study (1, 2, 5) of lungs from fatal cases has shown the combination of interstitial cellular infiltrations (lymphocytes, macrophages, plasma cells, and bizarre foreign body giant cells and inclusions) and diffuse patchy emphysematous alterations in alveolar structure. Right heart failure and degenerative changes in the liver have also been observed. Recovery of beryllium in most of the or-

[1] This work is part of a cooperative study of beryllium toxicology in human subjects carried out by the Department of Medicine of The University of Rochester School of Medicine and Dentistry under the direction of Dr. William S. McCann in collaboration with the Division of Medical Services of the Atomic Energy Project and aided by grants from the Lovejoy and Hochstetter Funds, University of Rochester School of Medicine and Dentistry.

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gans of a fatal case has completed the evidence for the specific etiology of granulomatosis in human subjects (9).

Observations on the cardio-respiratory functions[5] of 6 patients[6] with chronic delayed pulmonary granulomatosis due to beryllium are reported here (table 1 for physical data). The cases are arranged in order of increasing severity of disease from borderline disturbances to total disability. Four of these patients had known contacts with beryllium compounds under industrial working conditions. The other 2 patients were "neighborhood cases" since they did not work in the industry, but had contact with beryllium compounds only from atmospheric contamination in the close proximity to a manufacturing plant. The duration of respiratory distress in these 6 patients ranged between one and six years. The etiology was presumed in most by exclusion of all other differential diagnoses

TABLE 1

Physical data on patients studied

PATIENT	AGE	SEX	HEIGHT	WEIGHT	S. AREA	AP DIAMETER	RADIOLOGICAL CHEST VOLUME	TRANSVERSE CARDIAC RATIO
			cm.	*Kg.*	*sq. m.*	*cm.*	*liters*	
A. B.	26	M	169	56.0	1.65	17.5	9.22	0.446
M. C.	40	F	157	58.3	1.57	21.0	15.85	0.470
M. E. T.	23	F	167	59.9	1.66	20.0	11.14	0.384
M. H. C.	28	F	165	52.6	1.54	18.7	7.97	0.422
J. J.	31	M	162	52.8	1.54	21.0	8.83	0.524
W. R.	35	M	183	51.8	1.68	24.0	15.24	0.464

and was established in the third patient by identification of trace amounts of beryllium in the urine. In the sixth patient beryllium was recovered from kidney stones.[7]

METHODS

These patients were hospitalized for periods of observation ranging between two weeks and three months under complete metabolic control at all times. Except where indicated, all the measurements reported were obtained during nonfasting conditions. *Maximum breathing capacity* was measured by means of voluntary maximal hyperventilation for 30 seconds with the patient standing, using a mouth piece, high velocity valve, and a Douglas bag. The gas volume was measured in a standard Tissot spirometer, corrected to 37° C. and recorded per square meter of body surface area. Predicted values for both maximum breathing capacity and lung volumes were based on regression formulae derived by the investigators in the Chest Service of Bellevue Hospital (10). *Recumbent lung volumes* were determined in duplicate by means of the helium dilution technique (11). All partitions of the total capacity were corrected to 37° C. *Dead space* measurements were obtained and corrected to 37° C. by breathing data obtained from a modified A-13 Air Force face mask

[5] The metabolic studies on these patients will be the subject of a separate report.

[6] We are indebted to Dr. Joseph DeNardi of Lorain, Ohio, and Dr. Gordon Richardson of Towanda, Pennsylvania, for aiding in the arrangements in studying these patients.

[7] We are indebted to Dr. L. T. Steadman of the Department of Radiation Biology for making these measurements.

and continuous gas analyzer (12). *Bronchospirometry* (13) was performed with patients breathing oxygen and, when feasible, leg exercises were carried out during part of this procedure. Measurements of *alveolar pressure-velocity* relationships were obtained[8]. *Blood gas data* were based on arterial and mixed venous blood samples. Syringes containing 5 per cent sodium fluoride in heparin were routinely employed. Under sterile 1 per cent procaine infiltrative anesthesia, blood was obtained from either radial or femoral artery, and by means of *catheterization* of the right heart by Cournand's technique (14). Attempts were made to obtain respiratory data simultaneously for calculations of alveolar arterial gradients (employing Riley's method of calculating alveolar pO_2) (15) as well as cardiac output by direct Fick principle. The effects of mild exercise with the legs and of breathing pure oxygen were also noted. In one patient, who developed congestive heart failure, the initial effect of intravenous *digoxin* therapy were observed during a second catheterization, and in another the initial effects of intravenous pyrogen-free cytochrome C[9] were studied. The alterations of performance and of blood gas content from the ingestion of either *sodium chloride* or *ammonium chloride* were investigated in selected patients. Blood oxygen and carbon dioxide contents and capacities were measured by the Van Slyke (16) and Roughton-Scholander techniques (17), and gas tensions by the micro method of Riley, *et al.* (15).

In additon to the above, cardiac studies included serial *electrocardiograms* with complete chest leads (CF 1–6), *ballistocardiograms* (18), *pneumocardiograms* (19) and *intracardiac pressure-pulse* recordings employing a Statham Pressure Transmitter (20). *Venous pressure* was measured with a saline manometer, using 5 cm. below the sternum during recumbency as the zero reference point. *Circulation times* were measured simultaneously with a mixture of two drops of U.S.P. ether for arm-to-lung time, and 2 ml. of Macosol for arm-to-mouth time.

Exercise studies (21) were performed according to standardized routine of 10 minute periods of *rest, walking* on a motor-driven treadmill, and *recovery*. The rate of walking was either moderate 2.6 mph, or slow 1.7 mph (8 and 12 rpm or 46.7 and 70 meters per minute, respectively) and the grade of incline was either 0 per cent, 5 per cent, or 10 per cent. Simultaneous measurements at minute intervals were made of the heart rate, blood pressure, chest lead electrocardiogram, ventilation volume, respiratory rate, expired and "mid-capacity"[10] (rather than "alveolar") oxygen tension and carbon dioxide concentration. From these primary data, ventilation indices, oxygen consumption and debt, respiratory quotients, dead space volumes, etcetera, were calculated. All these gas volume data were corrected to square meter of body surface area at 0°C., 760 mm. mercury dry gas for ease of comparison between individuals. The appearance of symptoms of distress were recorded in relation to the other data. In 2 patients and one control, repeated exercise studies (as many as 32) were made to ascertain the range of variability in the several measurements listed above during exercise.

RESULTS

Patients with mild symptoms may show almost no physical signs of disease, yet exhibit characteristic changes on roentgenography of the chest. Functional studies reveal appreciable deviations from normal performance that account for the exertional dyspnea and intolerance of more than moderate activity.

[8] We are indebted to Dr. Arthur Otis, Department of Physiology, for making these measurements.

[9] Obtained from Biorganic Laboratories, Inc.

[10] In the traditional sense, measurement of alveolar air involves determination of gas concentrations after *forced* expiration; "mid-capacity" concentrations refer to those obtained at the end of normal expiration. In exercise studies, only the "mid-capacity" can be used.

Cases

A. B., a 27 year old white married male, had industrial contact with irritating fumes from a beryllium alloy process for three months during 1940. He was in excellent health until 1944, when he developed bouts of chills, fever, irritative coughing, wheezing and dyspnea on exertion while in the Army. Chest roentgenographic examination was reported negative at that time, but by October, 1947, symptoms had progressed, weight loss from anorexia had occurred, and another chest roentgenogram revealed diffuse bilateral lesions compatible with beryllium granulomatosis. He has been unable to work since that time. On examination in March, 1948, vital signs were normal except for slight tachypnea. The patient appeared slender; he was using his accessory muscles of respiration even for quiet breathing. There was neither cyanosis nor clubbing of the fingers and the chest examination was otherwise unremarkable except for accentuation of breath sounds over the left lower lobe of the lung. A roentgenogram of the chest showed enlarged hilar root shadows with soft outlines and diffuse nodularity throughout the lung fields (figure 1a). The patient was unable to remain for complete studies, but the following observations were made. The red blood count was 5,000,000 per cu. mm., the hemoglobin concentration was 22.3 Gm. per 100 cc. and the hematocrit was 51 per cent. The respiratory pattern was irregular, with sighing indicative of emotional disturbance, which was compatible with the observed behavior. His maximum breathing capacity was only 56 per cent of the predicted value and in some measure was low because of poor cooperation. Determination of lung volumes showed marked reduction of total capacity (table 2, figure 2) to 3.0 liters, or 58 per cent of the value predicted from the height formula. Measurement of chest volume on the roentgenogram was also reduced, suggesting inability to adequately expand the chest, and the vital capacity was limited to 2.0 liters recumbent, or 2.5 liters standing. The alveolar pressure-velocity measurements were normal (table 2). The electrocardiogram, the ballistocardiogram, the venous pressure, and circulation times, were all normal (table 3). The patient was able to perform standard exercise tests (at rate of 2.6 mph) at inclines of 0, 5, and 10 per cent grades for 10 minutes (table 4, figure 5). On the latter two, he experienced severe dyspnea, sweating and substernal distress. His legs became very tired. There was a paradoxical pulse with average heart rates up to 134 per minute. No electrocardiographic changes were observed, and the oxygen debts were normal. The ventilation indices were but slightly increased, and the pulmonary efficiency in relation to work was almost normal (figure 3).

This patient's reduced ventilatory capacity and total lung volume were in contrast to his apparent inability to hyperventilate during rest or exercise as much as other patients (figure 3). Possibly the secondary polycythemia accounted for this. Unfortunately no arterial blood samples were obtained in order to evaluate either the alveolar oxygen gradient or the degree of hypoxemia. In an older individual, relatively the same degree of granulomatous change may be associated with greater functional impairment, as indicated by the next case.

M. C., a 40 year old white woman, developed a contact dermatitis on both arms and hands as a result of an industrial exposure to beryllium powders for three months during 1944. She discontinued this work and was in good health until August, 1946, when she began to notice coughing while on vacation in Oregon. Exertional dyspnea was first noted in June, 1947; a chest roentgenogram obtained a few months later revealed "cloudy" changes compatible with "beryllium poisoning." She experienced no anorexia, and weight increased from 128 to 136 pounds during the last six months of 1947. On examination in April, 1948, vital signs were normal, and the patient appeared well. Frequently she exhibited an irritative coughing, put showed no tachypnea, dyspnea, cyanosis, limitation of chest expansion, or clubbing of the fingers. Occasionally wheezes and rhonchi were audible on

auscultation of the lungs. The heart was not enlarged, the rhythm was regular, and the sounds were of good quality, although the second pulmonic sound was accentuated. Roentgenographic examination of the chest showed moderately fine, confluent, soft nodulations scattered diffusely through both lung fields (figure 1b). (Metabolic studies revealed no abnormalities except for slight lowering of plasma albumin to 3.3 Gm. per 100 cc. as determined by electrophoresis. Because of a tendency towards rouleaux formation, the red blood cell count was 6.76 millions per cu. mm., but the contentration of hemoglobin was only 11.8 Gm. per 100 cc.) The maximum breathing capacity was 97 per cent of the expected

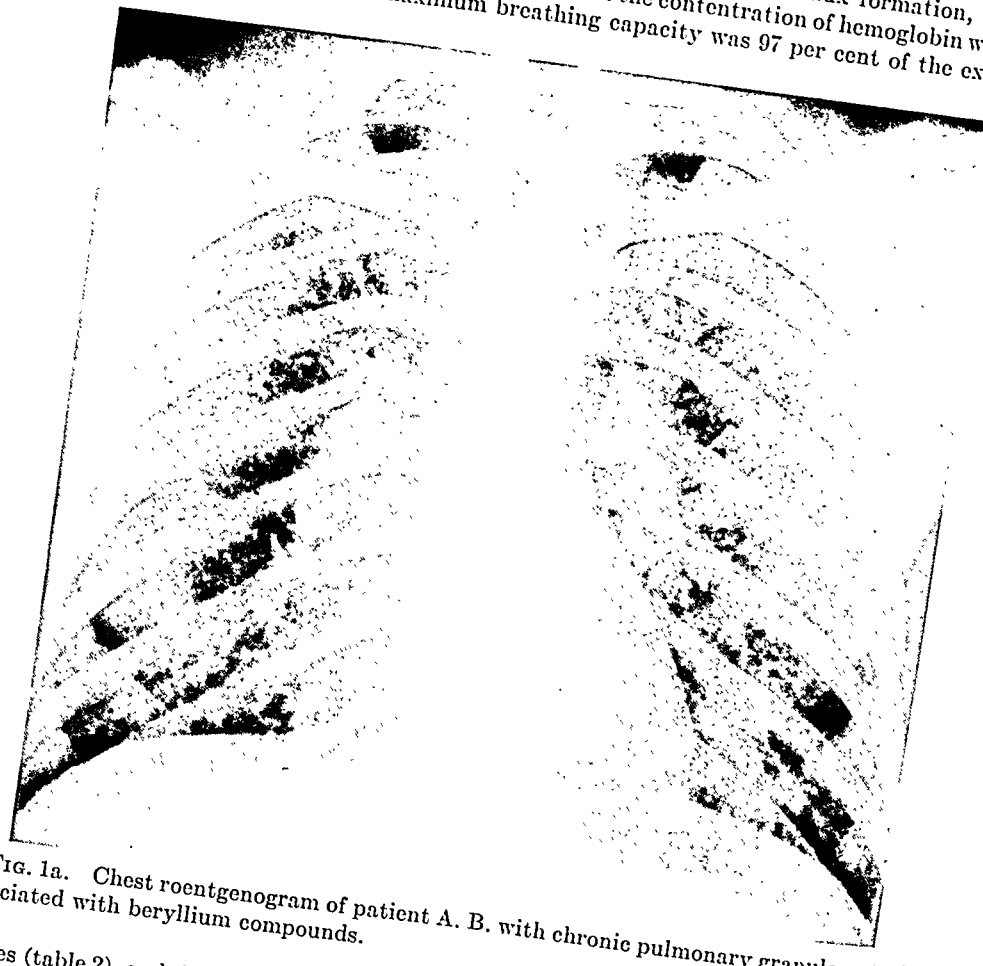


FIG. 1a. Chest roentgenogram of patient A. B. with chronic pulmonary granulomatosis associated with beryllium compounds.

values (table 2), and the breath-holding time was 34 seconds. The vital capacity and total pulmonary capacity were reduced to 1.87 and 3.21 liters, respectively, or 67 per cent and 89 per cent of the predicted values (table 2, figure 2).

Bronchospirometry while breathing oxygen showed that the proportion of ventilation between the two lungs was normal, but that 67 per cent of the oxygen uptake was performed by the right lung. The electrocardiogram was normal, as were the venous pressure and the arm-to-mouth circulation time, but the arm-to-lung circulation time was prolonged to 10 seconds (table 3). By right heart catheterization, the right ventricular pulse pressure was 25 to 37 mm. of mercury during rest, and increased to 45 to 67 mm. of mercury during mild leg exercises. The resting cardiac output was normal: 4.7 liters per minute by direct Fick principle, and increased to 6.65 liters during exercise. Blood gas studies showed a resting

arterial oxygen saturation of 86 per cent, hemoglobin concentration of 12.46 Gm. per 100 cc., and pO_2 of 75 mm. The alveolar-arterial oxygen gradient was not greatly increased, and the resting level of ventilation was a high normal value despite the presence of borderline anemia and definite hypoxemia (figure 4). Exercise studies showed that the patient was able to walk (at the rate of 2.6 mph) on the level without symptoms, experienced dyspnea at 5 per cent grade of incline, and was unable to walk more than five minutes at 10 per cent grade because of more intense dyspnea. The ventilation indices were moderately increased whereas the oxygen debts were normal at 0 and 5 per cent grades, but were in-

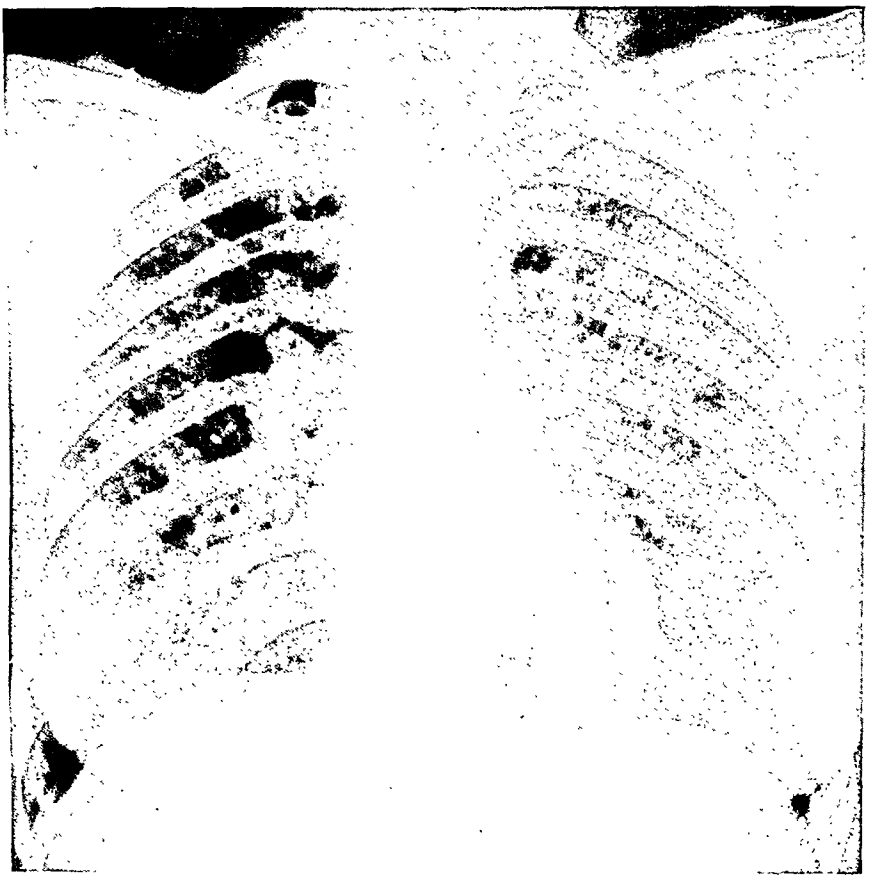


FIG. 1b. Chest roentgenogram of patient M. C. with chronic pulmonary granulomatosis associated with beryllium compounds.

creased markedly to 54 per cent at 10 per cent grade walking (table 4). The pulmonary efficiency in relation to work (figure 3) was appreciably reduced.

This patient, M. C., also had moderate reduction in vital capacity, yet in contrast to A. B. was able to maintain a normal maximum breathing capacity. Her pulmonary efficiency was impaired because of moderate increase in alveolar-arterial gradient which prevented normal saturation of arterial blood with oxygen. With increased arm-to-lung circulation time and pulmonary hypertension during exercise, she presented evidence of pulmonary resistance and right heart strain (noted by accentuation of the second pulmonic sound). Under

ordinary activity the exertional dyspnea was due to pulmonary insufficiency only, whereas with more strenuous exertion the unusually large oxygen debt indicated acute circulatory insufficiency and possible incipient heart failure.

Another of these patients experienced attacks of dizziness in addition to exertional dyspnea as an aftermath of diffuse pneumonitis.

M. E. T., a 23 year old white woman, had intermittent exposure to atmospheric contamination with dust containing beryllium compounds for an indefinite period during pregnancy

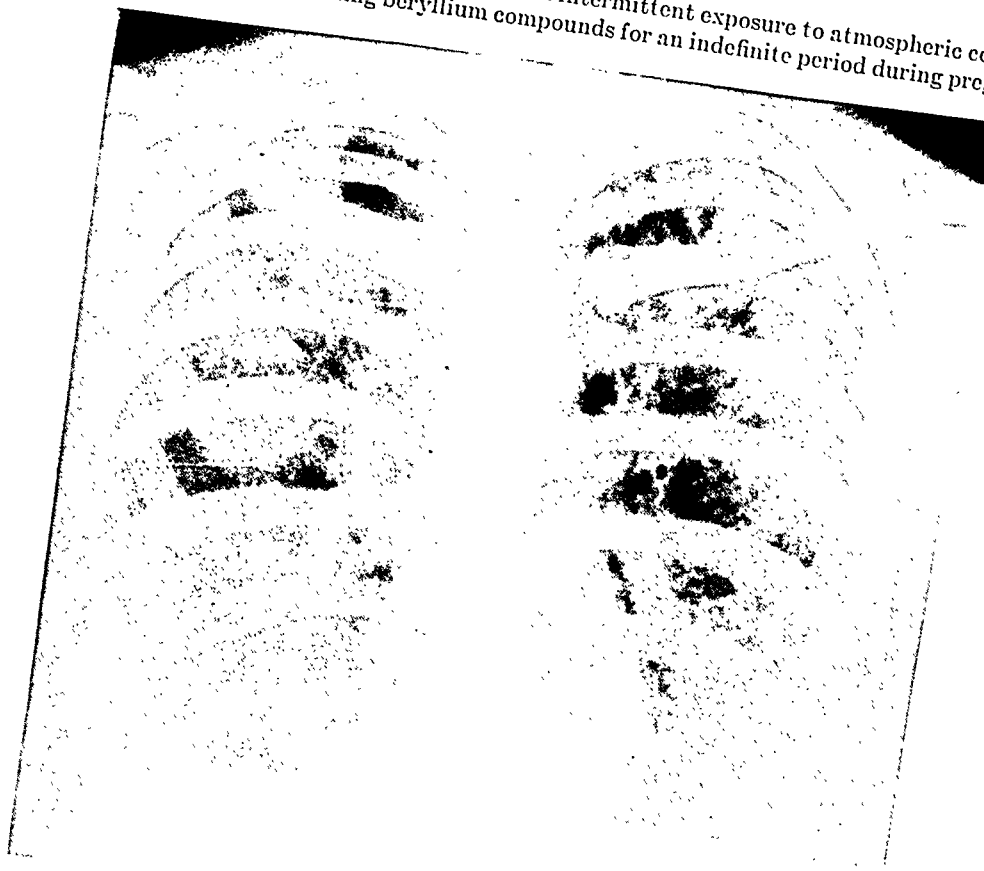


FIG. 1c. Chest roentgenogram of patient M. E. T. with chronic pulmonary granulomatosis associated with beryllium compounds.

in 1946. Following this, the patient lived with her parents adjacent to a plant manufacturing beryllium products. Fumes from this factory caused pharyngeal irritation and coughing, especially on "damp and muggy days." By March, 1947, she noted exertional dyspnea, coughing productive of mucous, substernal tightness in chest, and occasionally nausea. Later when she developed chills, fever and roentgenographic evidence suggestive of miliary tuberculosis, she was hospitalized in a sanatorium. She exhibited a febrile course, required continuous oxygen therapy for dyspnea and cyanosis, and was treated with both penicillin and streptomycin for a few days. She gradually recovered, became ambulatory, and was observed here from December, 1947, to February, 1948. On admission to Strong Memorial Hospital she appeared almost well, but there was cyanosis of the nails. Chest expansion

was limited and inconstant rales were audible over the base of each lung. Roentgenographic examination of the chest showed diffuse pulmonary haziness, reticular markings and minute nodulations; the heart was not enlarged (figure 1c). With exertion the patient became dyspneic and complained of dizziness. During the period of observation the intensity of these two symptoms fluctuated considerably, and at times she became easily fatigued and exhibited orthopnea. Some of the variation in symptomatology was related to emotional factors represented by frequent sighing respirations and a tendency to hyperventilate. The routine hospital laboratory data were normal, except for reduction in plasma albumin concentration to 3.3 Gm. per 100 cc., as determined by electrophoresis, and diminished blood volume by the Evans blue technique[11]. The patient's predicted blood

FIG. 1d. Chest roentgenogram of patient M. H. C. with chronic pulmonary granulomatosis associated with beryllium compounds.

volume was 4,050 ml., but the observed value was 2,960 ml. with an hematocrit of 44.5 per cent and plasma volume of 1,650 ml. The maximum breathing capacity was 53 per cent of the predicted value (table 2). Alveolar pressure-velocity relationships were normal. Determination of lung volumes showed a moderate reduction of both the total pulmonary capacity and the vital capacity (table 2, figure 2). In an attempt to evaluate further the dyspnea and dizziness by altering her electrolyte balance, the patient was given a course of sodium chloride 2.0 Gm. orally, four times daily, for four days prior to the expected onset of

[11] We are indebted to Dr. L. A. Kohn for these data.

menses. Serial measurements of lung volumes showed that the total and vital capacities diminished appreciably. She complained of greater exertional dyspnea and orthopnea, together with diminished exercise tolerance. The venous pressure was 11 cm. of water, and rose to 12 cm. with pressure over the right upper quadrant of the abdomen. There was no significant change in basal weight from day to day. The observed blood volume increased to 3,600 ml. with an hematocrit of 41.8 per cent and a plasma volume of 2,100 ml. Coincident with the onset of the menstruation, the patient was then given 2.0 Gm. of ammonium chloride orally, three times daily, instead of salt. The basal weight declined 1.0 Kg., the exertional dyspnea diminished, but the intensity of dizziness became so severe that the ammonium chloride had to be discontinued after forty-eight hours. Concomitantly there

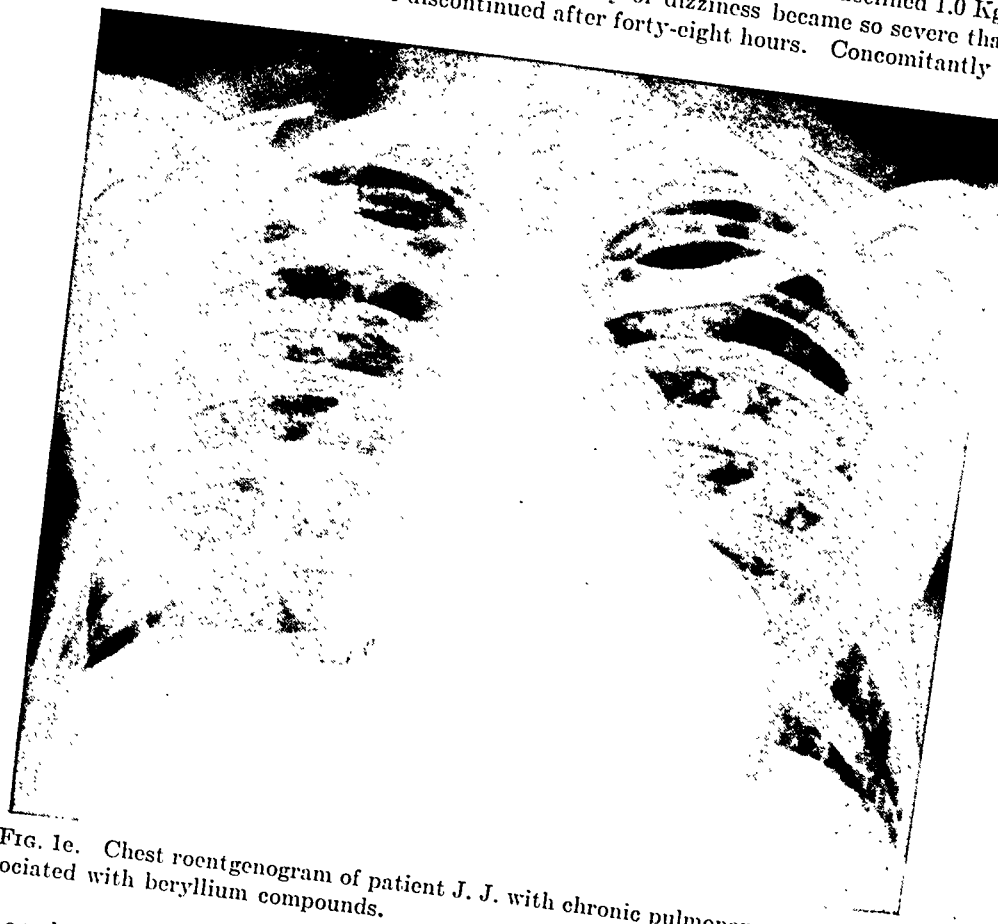


FIG. 1e. Chest roentgenogram of patient J. J. with chronic pulmonary granulomatosis associated with beryllium compounds.

was a reduction in the arterial pCO_2 from 44.5 mm. to 36.5 mm., and the residual air volume declined to 410 ml. or 13.5 per cent of the total capacity. The arm-to-lung circulation time was prolonged to 14 seconds. The performance of exercise caused intense substernal pain during this period, although no electrocardiographic alterations were observed. Less marked changes were observed when the ammonium chloride was repeated one week later, after the completion of the menses.

Right heart catheterization showed a normal resting right ventricular pulse pressure of 16 mm. of mercury. The arterial blood was 87 per cent saturated with oxygen and the pO_2 was 72 mm. Repeated exercise tolerance studies at 0, 5, and 10 per cent grades of incline showed considerable daily fluctuation in intensity of dyspnea and dizziness, as well as endurance. Patient was able to walk only seven minutes at 5 per cent grade, and four

minutes at 10 per cent grade because of dyspnea and dizziness. Although the ventilation indices were increased, the oxygen debts were normal, except for the 10 per cent grade when it was 22 per cent, largely because of the shortened exercise period. In a series of exercise tests at 0 per cent grade, analysis of the coefficients of variability of the several measure-

FIG. 1f. Chest roentgenogram of patient W. R. with chronic pulmonary granulomatosis associated with beryllium compounds.

ments obtained during exercise showed the oxygen consumption to be the most variable (5.7 per cent) whereas the rate of oxygen absorption per liter of air ventilated was the least variable (2.4 per cent). Differences in anxiety, rather than in gaseous diffusion, therefore, seemed to account for the variations in the patient's symptoms from day to day.

This patient's exertional dyspnea also was related to reduction in ventilatory capacity and complementary lung volume, in addition to the hyperventilation

compensating for hypoxemia. There was no evidence of right ventricular strain under ordinary circumstances. The dizziness may have been in part related to

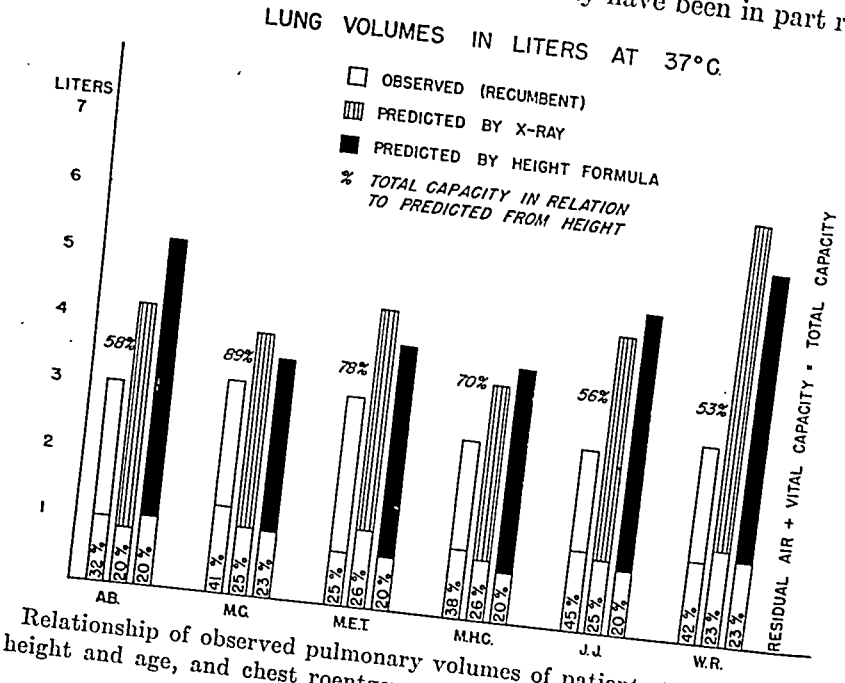


FIG. 2. Relationship of observed pulmonary volumes of patients to predicted values based on height and age, and chest roentgenograms.

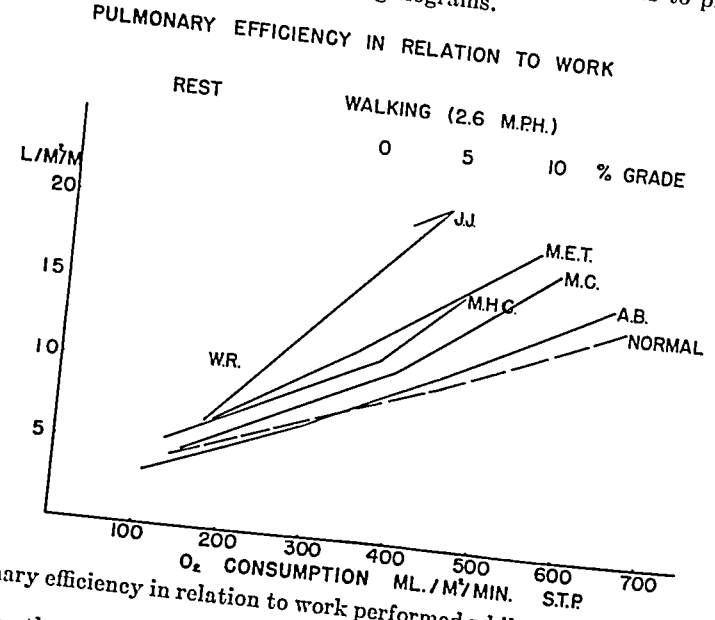


Fig. 3. Pulmonary efficiency in relation to work performed while walking on the treadmill.

hypocapnia, for the ingestion of ammonium chloride during the menstrual period was associated with a reduction in arterial pCO_2. Since the residual air volume simultaneously was markedly reduced, the buffering action of this lung volume

partition may have been sufficiently altered to permit greater escape of carbon dioxide without significant increases in ventilation. The mechanism of this reversible action is difficult to explain, since it was less marked when repeated after menstruation, but may have been related to pulmonary venous vasoconstriction. Finally, the measurement which reflected the diffusion of oxygen into the blood during exercise was the least variable value, and showed no correlation with the patient's variations in symptoms. The fourth patient in this series

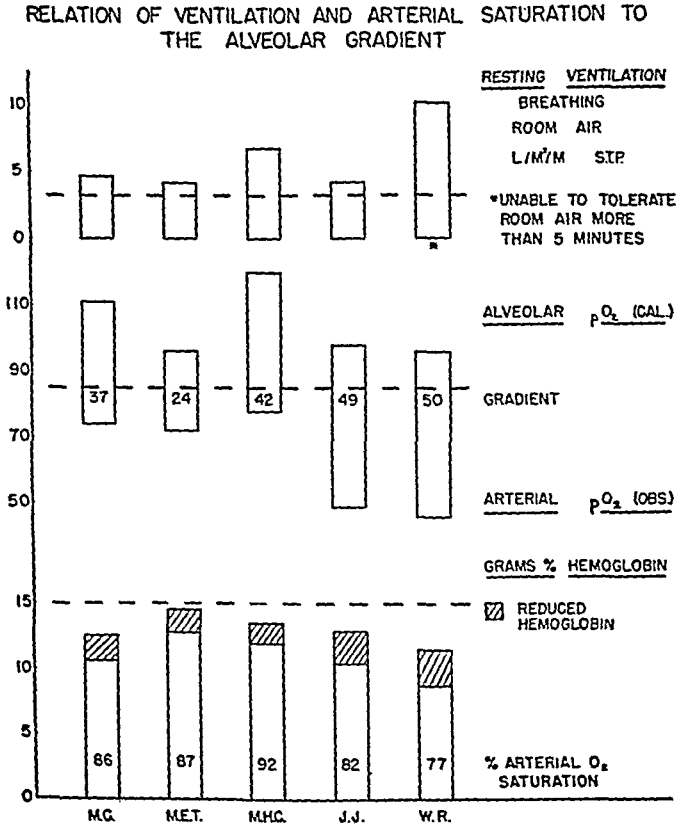
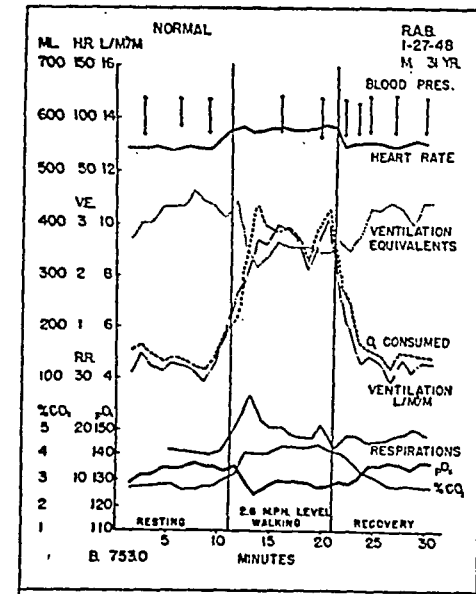
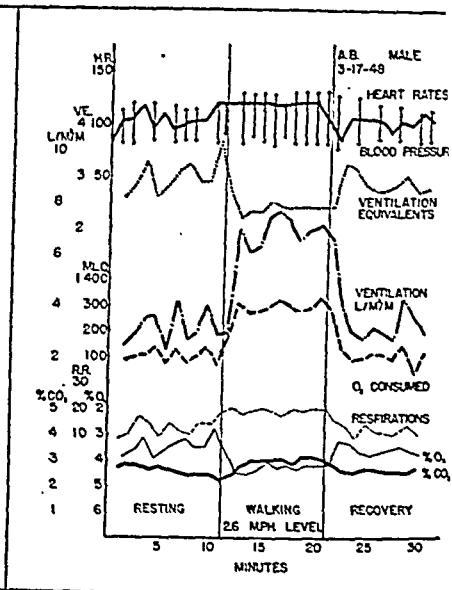


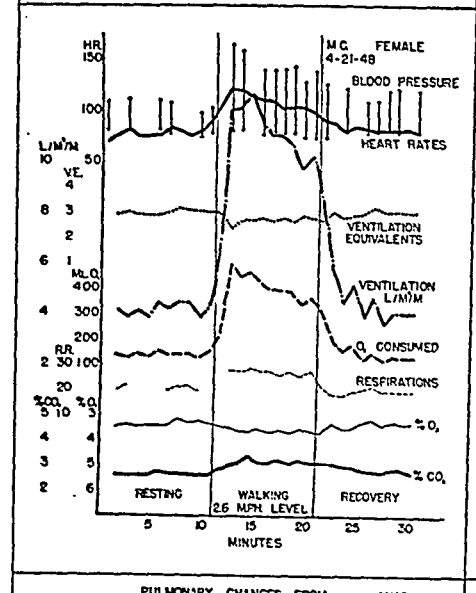
FIG. 4. Relation of ventilation and arterial oxygen saturation, at rest, to alveolar-arterial oxygen gradient.

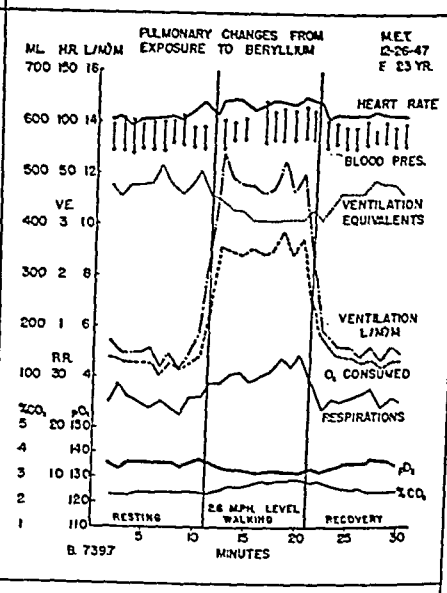
came from the same neighborhood as the third, but her manifestations of chronic pulmonary granulomatosis were somewhat different at the time of observation.

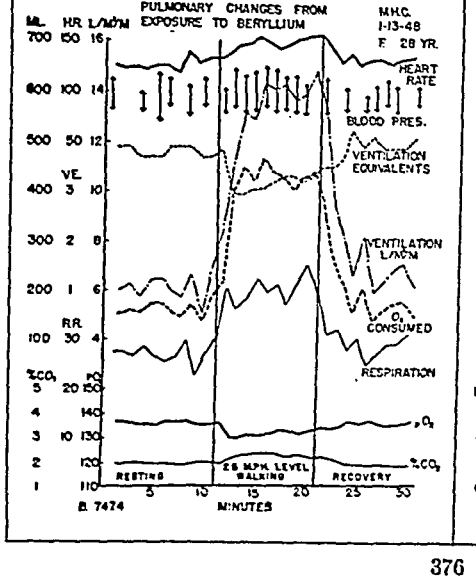
M. H. C., a 28 year old white married woman, also lived in close proximity to the same beryllium factory (as did patient M. E. T.) from 1944 to 1948. In May, 1946, she noticed coughing which was occasionally productive of blood-streaked sputum. Inhalation of fumes from this factory were known to be irritating to her. The paroxysms of coughing gradually became more severe, and by July, 1947, she had recurrent chills, fever, orthopnea, and dyspnea at night as well as on exertion. Because of anorexia she lost 28 pounds in weight. Cyanosis became prominent on exertion, and in November, 1947, she moved to another location and her symptoms began to subside. On examination in January, 1948, she exhibited moderate respiratory distress, limitation of chest expansion, mild cyanosis,

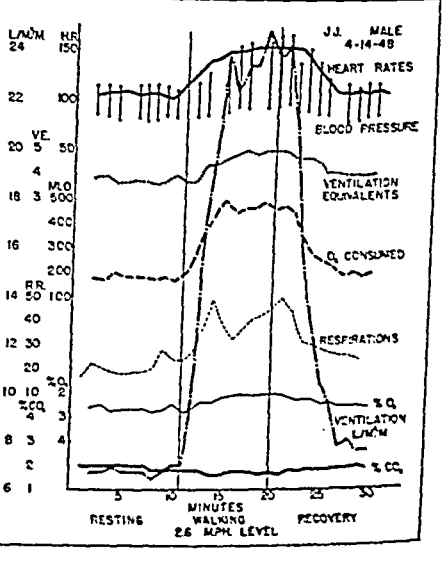

NORMAL
R.A.B.
1-27-48
M 31 YR.
ML. HR L/MIN
BLOOD PRES.
HEART RATE
VENTILATION EQUIVALENTS
O2 CONSUMED
VENTILATION L/MIN
RESPIRATIONS
RESTING
2.6 MPH LEVEL WALKING
RECOVERY
B. 7530
MINUTES


A.B. MALE
3-17-48
HEART RATES
BLOOD PRESSUR
VENTILATION EQUIVALENTS
VENTILATION L/MIN
O2 CONSUMED
RESPIRATIONS
RESTING
WALKING 2.6 MPH LEVEL
RECOVERY
MINUTES


M.G. FEMALE
4-21-48
BLOOD PRESSURE
HEART RATES
VENTILATION EQUIVALENTS
VENTILATION L/MIN
O2 CONSUMED
RESPIRATIONS
RESTING
WALKING 2.6 MPH LEVEL
RECOVERY
MINUTES


PULMONARY CHANGES FROM EXPOSURE TO BERYLLIUM
M.E.T.
12-26-47
F 23 YR.
ML. HR L/MIN
HEART RATE
BLOOD PRES.
VENTILATION EQUIVALENTS
VENTILATION L/MIN
O2 CONSUMED
RESPIRATIONS
RESTING
2.6 MPH LEVEL WALKING
RECOVERY
B. 7397
MINUTES


PULMONARY CHANGES FROM EXPOSURE TO BERYLLIUM
M.H.C.
1-13-48
F 28 YR.
ML. HR L/MIN
HEART RATE
BLOOD PRES.
VENTILATION EQUIVALENTS
VENTILATION L/MIN
O2 CONSUMED
RESPIRATION
RESTING
2.6 MPH LEVEL WALKING
RECOVERY
B. 7474
MINUTES


J.J. MALE
4-14-48
HEART RATES
BLOOD PRESSURE
VENTILATION EQUIVALENTS
O2 CONSUMED
RESPIRATIONS
VENTILATION L/MIN
MINUTES
RESTING
WALKING 2.6 MPH LEVEL
RECOVERY

and slight clubbing of the fingers. There was a sinus tachycardia, averaging around 120 per minute, and the blood pressure was reduced to 100/70 mm. of mercury. Rales were audible over the lung bases. Roentgenographic examination of the chest showed diffuse granular infiltrations throughout the lung fields (figure 1d). The maximum breathing capacity was 43.5 liters per square meter, or 75 per cent of the predicted value (table 2). Breath-holding time was 23 seconds. Alveolar pressure-velocity relationships were normal.

TABLE 2

Maximum breathing capacity, lung volumes, and physiological dead spaces in liters at 37° C. and alveolar pressure at 500 ml/sec. velocity air flow

PATIENT	MAXIMUM BREATHING CAPACITY			VITAL CAPACITY	COMPLEMENTARY AIR	RESERVE AIR	MID-CAPACITY	RESIDUAL AIR	TOTAL CAPACITY	M.C.	R.A.	TIDAL	DEAD SPACE	ALVEOLAR* PRESSURE
	Obs.	Pred.	Per cent							per cent	per cent			mm.
A. B.	41.0	73.0	56	2.05	1.11	0.71	1.69	0.98	3.03	56	32	0.62	—	13.5
M. C.	50.7	52.4	97	1.87	1.19	0.65	1.99	1.34	3.21	62	41	0.51	—	—
M. E. T.	31.9	60.4	53	2.34	1.46	0.52	1.31	0.79	3.13	42	25	0.50	0.161	12.0
M. H. C.	43.5	58.0	75	1.67	0.71	0.51	1.54	1.03	2.70	57	38	0.67	0.155	12.0
J. J.	51.5	70.0	74	1.51	1.27	0.76	1.46	1.23	2.74	53	45	0.47	0.120	—
W. R.	10.2	68.0	15	1.74	1.60	0.78	2.04	1.26	3.00	67	42	0.86	—	9.0

* Mean normal pressure in 30 normals is 11.3 mm. ± 3.43[3].

TABLE 3

Cardiovascular measurements at rest

PATIENT	VENOUS PRESSURE, CM. OF WATER	CIRCULATION TIME		RIGHT AURICULAR PRESSURE	RIGHT VENTRICULAR PRESSURE	CARDIAC OUTPUT	STROKE VOLUME	BALLISTOCARDIOGRAM
		A-L	A-M					
		sec.	sec.			L/min.	ml.	
A. B.	9.4	7	15	—	—	—	—	Normal
M. C.	8.4	10	15	5	25	4.7	58	Borderline
M. E. T.	9.5	8.8	15	—	12	—	—	Low amplitude
M. H. C.	8.1	7	14	—	25	—	—	Shallow I, deep K Low amplitude
J. J.	13.5	11	16	13	78	3.09	28	Borderline normal
W. R.	−1.0	7.5	14	30	65	5.92*	70	Abnormal (marked respiratory variations

* Breathing oxygen.

Determination of lung volumes revealed that the vital capacity was reduced to 1.67 liters or 54 per cent of the predicted value (figure 2), and a less marked reduction in total capacity was

FIG. 5. Exercise performance of one normal control (R. A. B.) and 5 patients in order of increasing cardio-respiratory disability.

Top row, left to right: R. A. B. (control) and patient A. B.; Second row, left to right: M. C. and M. E. T.; and Bottom row, left to right: M. H. C. and J. J. (Note paradoxical responses in J. J.).

observed. Except for sinus tachycardia and depression of the ST segment in lead 1, the electrocardiogram was unremarkable. Venous pressure, circulation times, and blood pressure were normal, but the right ventricular pulse pressure at rest was near the upper limits of normal (table 3). Despite the low vital capacity the arterial blood was 92 per cent satu-

TABLE 4

Exercise responses to treadmill walking at 2.6 mph

PATIENT	ENDURANCE	VENTILATORY EQUIVALENT*		VENTILATION INDEX**	OXYGEN DEBT	HEART RATE*	BLOOD PRESSURE*	VENTILATION*	O_2 CONSUMPTION	R.Q.*	SYMPTOMS AND SIGNS
		Rest	Exercise								
	min.				*per cent*		*mm.Hg*	*L/M²/min.*	*ml./M²/min.*		
0 per cent Incline											
A. B.....	10	3.00	2.38	8.16	7	120	130/81	6.82	286	0.71	Slight dyspnea
M. C.....	10	2.99	2.75	10.3	9	110	145/77	10.81	395	0.83	None
M. E. T.	10	3.90	3.34	13.8	10	125	118/87	11.78	352	0.92	Slight dyspnea
M. H. C.	10	3.80	3.01	19.8	15	143	129/85	11.35	376	—	Dyspnea, fatigue
J. J.....	9	3.79	4.72	38.2	30	108	117/83	20.96	439	0.83	Severe dyspnea, cough, fatigue
5 per cent Incline											
A. B.....	10	3.00	2.36	14.0	16	124	131/79	10.64	451	0.83	Severe dyspnea, sweating, substernal distress
M. C.....	10	3.06	2.95	16.5	16	131	—	14.67	506	—	Dyspnea, fatigue
M. E. T.....	7	3.50	3.40	16.8	9	133	—	15.13	445	0.78	No comments
M. H. C.....	7	4.40	3.33	21.5	16	150	140/77	15.40	464	—	Dyspnea, left chest pain
J. J..... (1.7 mph).	10	4.48	4.88	36.2	27	141	116/74	19.65	398	1.07	Low voltage ECG
10 per cent Incline											
A. B.....	10	3.20	2.47	16.5	12	134	154/81	15.89	641	0.83	Severe dyspnea, sweating, legs tired, paradoxical pulse
M. C.....	5.16	3.08	3.07	25.0	54	148	161/76	17.60	574	0.98	Dyspnea, sweating
M. E. T.....	4.5	3.50	3.31	20.3	22	136	117/71	18.15	548	0.93	Dyspnea, dizziness

* Average value per minute for exercise period.

** Mean ventilation index in 35 normals is 6.12 ± 2.08.

rated, and the pO_2 was 78 mm. Exercise tolerance studies demon strated that she was able to walk at 2.6 mph and at 0 and 5 per cent grades, but that fatigue and dyspnea developed even with the least exertion. Her tolerance was shortened by grade walking to seven minutes, and she was unable to walk at 10 per cent grade without supplemental oxygen. The ventilation indices were greatly elevated to 19.8 and 21.5 at 0 and 5 per cent grades, re-

spectively, showing the extent of the hyperventilation which prevented more serious degrees of hypoxemia (table 5). Pulmonary efficiency in relation to work diminished as soon as grade walking was attempted (figure 3). The least variable factor during exercise, based upon coefficients of variability of multiple tests, was the rate of oxygen absorption per liter of ventilation (5.3 per cent), whereas ventilation volume exhibited the greatest variation (11 per cent). The oxygen debts were normal, yet clinically the patient often exhibited marked accentuation of second pulmonic sound, transient gallop rhythm, left chest pain, sweating, and exhaustion as a result of this exertion. She presented no signs of heart failure, and the administration of 1.5 mg. of digoxin orally in two divided doses caused anorexia and nausea but failed to diminish intensity of exertional dyspnea or to improve exercise tolerance or performance. Ammonium chloride 2 Gm. orally, three times daily, for two days induced anorexia and dizziness, but no other changes similar to those in patient M. E. T. were observed.

This patient, M. H. C., differed from the preceding one by more intense exertional dyspnea because of a smaller vital capacity. Her maximum breathing capacity was appreciably greater, and this may account for the more nearly normal level of arterial oxygen saturation. During the period of observation there was an appreciable increase in vital capacity together with a slowing of the heart rate at rest and exercise. The exercise tolerance was not as good, however, probably because of pulmonary hypertension during exertion. In contrast to this patient is the disability from advanced cor pulmonale exhibited by the fifth and sixth patients, both of whom had definite industrial exposures to beryllium compounds.

J. J., 31 years of age, worked for two months as a sifter of fluorescent powders containing beryllium. Despite protective clothing and face mask, he had to discontinue this work in December, 1942, because of coughing and exertional dyspnea. Roentgenographic examination of the chest was reported to show "dust in the lungs." After these symptoms subsided he returned to work on another job which exposed him to fumes of various acids and ammonia, as well as beryllium, tungsten, and carborundum dusts on various occasions. Early in 1945, he had a recurrence of coughing, exertional dyspnea, together with fatigue, anorexia, weight loss, night sweats, chills and fever which progressed over three months. Paroxysmal nocturnal dyspnea and orthopnea were also noted. He was admitted to Trudeau Sanatorium where exhaustive diagnostic and functional studies[12] were made. The arterial oxygen saturation was 86 per cent and the total lung volume was 2.73 liters with 24 per cent residual air content. The exercise tolerance was limited to 2.5 mph level walking for 5 minutes, and the ventilatory equivalent for oxygen was 4.7 liters per 100 ml. of oxygen per minute. During the next year and one-half the patient improved somewhat, but lost 40 pounds in weight. Tubercle bacilli were never demonstrated in the sputum and a tuberculin skin test was negative to 10 mg. Examination of material obtained from biopsy of skin lesions on the hand showed lesions with noncaseous central necrosis which were interpreted as "changes characteristic of Boeck's sarcoid." Minimal symptomatic relief was obtained from quinine and neoarsphenamine therapy. When the patient was returned home, he gradually improved, gained weight from 117 to 133 pounds and by the summer of 1947 was able to mow part of his lawn. After that time, clubbing of the fingers, accentuation of the second pulmonic heart sound, more exertional dyspnea, cyanosis, and weight loss indicated further hypoxemia and cor pulmonale. Examination in April, 1948, revealed a chronically ill, malnourished white male, with obvious respiratory distress even at rest. There was tachypnea of 24 per minute at rest, cyanosis, marked clubbing of the nails, limited chest expansion and

[12] Functional studies were done by Dr. George Wright, Saranac Lake, New York.

utilization of the accessory muscles for respiration. The lungs were resonant to percussion, but rales were audible on examination of the bases. By percussion the pulmonary conus was prominent and the heart was enlarged to the right. The second pulmonic sound was accentuated and a systolic murmur was heard along the left sternal border. Generalized atrophy of the skeletal musculature and myotactile irritability confirmed the history of extensive loss of weight. Roentgenographic study of the chest showed soft infiltrations of both lung fields with increased bronchovascular markings. There was prominence of the right auricle and pulmonary conus (figure 1e). The red blood cell count was 5.5 million per cu. mm., and the hemoglobin concentration was 18.5 Gm. per 100 cc. (Sahli). The urine showed a one plus reaction for albumin, and the blood urea nitrogen was 27 mg. per 100 cc. Bromsulfthalein test showed 60 per cent retention at the end of forty-five minutes, after a dosage of 5 mg. per Kg. of body weight. Electrophoretic analysis of plasma protein was unremarkable save for slightly low albumin of 3.46 Gm. per 100 cc. and an increased concentration of alpha-2 globulin. A vitamin A absorption test was abnormal. The maximum breathing capacity was 51.5 liters per square meter, or 73.5 per cent of the predicted value (table 2). The lung volumes showed a very great reduction in vital capacity to 39 per cent, and total capacity to 56 per cent of the predicted values (table 2, and figure 2). The electrocardiogram was abnormal because of right axis deviation, and peaked P waves in lead II in addition to sinus tachycardia. The venous pressure was increased to 135 cm. and fell paradoxically to 11.5 cm. with pressure over the right upper quadrant of the abdomen. The arm-to-lung circulation time was prolonged to 11 seconds; arm-to-mouth time was 16 seconds. Venous catheterization of the right heart showed increased right auricular pressure (13 mm.) and right ventricular pulse pressure was as high as 78 mm. of mercury. The cardiac output by direct Fick principle was abnormally low or 3.1 liters per minute. Arterial oxygen saturation was depressed to 82 per cent with a pO_2 of 50 mm. Breathing pure oxygen increased these values to 95.6 per cent and 114 mm., respectively, but probably did not improve tissue utilization since the A-V oxygen difference fell from 9.4 to 7.78 vol. per cent. Exercise tolerance studies showed a marked impairment of pulmonary efficiency in relation to work, and this was aggravated by any increment over the resting value since exercise caused an increase in ventilatory equivalents (figures 3, 5, table 4). This unusual response was due to the extraordinary degree of hyperventilation induced by effort, and may also indicate further increments in pulmonary hypertension. The patient was unable to perform even 5 per cent grade walking except at the slow speed of 1.7 mph. Both ventilation indices and oxygen debts were abnormally high for either level or grade walking (table 4).

This fifth patient exhibited the late sequelae of chronic pulmonary granulomatosis due to beryllium, namely: contracted lungs with relative emphysema, cor pulmonale with incipient congestive failure, malnutrition, liver disease, and early renal disease in addition to severe anoxemia. Despite the latter, there was no marked polycythemia (figure 4). The ease with which such a patient may suffer serious complications is revealed by the following observations:

During the course of nitrogen balance studies, the patient was given 25 mg. of testosterone propionate intramuscularly daily for twelve days to ascertain its anabolic effects. The patient gradually became edematous, suffered more intense dyspnea, coughing and orthopnea even after the drug had been discontinued for a week. The laboratory evidence of congestive heart failure is presented in table 5. During the course of a second venous heart catheterization, the initial effects of 1.0 mg. of digoxin intravenously were recorded (figure 6). There was a transient lowering of right auricular pressure followed by an increase in right ventricular pulse pressure, and improvement in pressure pattern. Within forty minutes the right heart output improved sufficiently to overload the pulmonary circulation and pulmonary edema ensued. Fortunately the patient was promptly relieved by a phlebotomy

TABLE 5

Effects of congestive heart failure in patient J. J.

	APRIL 1948	MAY 1948	AFTER DIGOXIN THERAPY
Basal Weight, Kg	52.1	58.1	54.2
Lung volumes in liters			
Vital capacity	1.51	1.31	—
Residual air	1.23	1.50	—
Total capacity	2.74	2.81	—
Per cent residual air	45 per cent	53 per cent	—
Venous pressure			
cm. water	13.5	20.5	11
Circulation times, seconds			
Arm-to-lung	11	21	10
Arm-to-mouth	16	25	18
Resting metabolic rate	+19 per cent	+32 per cent	+43 per cent
Exercise tolerance			
1.7 mph, 0 per cent grade			
Endurance, minutes	10	8	10
Ventilation index	25	32	33
Oxygen debt	12 per cent	22 per cent	17 per cent
Average exercise			
Ventilatory equivalents	3.9	4.4	3.9
Heart rate	146	128	122
Blood pressure	141/79	118/83	143/77
Pulse pressure	62	35	66
Symptoms	Dyspnea	Severe dyspnea, exhaustion	Moderate dyspnea
Ballistocardiogram			
Pattern	Prominent H	Irregular	—
Amplitude	fair	low	—
Arterial Blood			
pCO_2, mm	41	34	—
pO_2, mm	50	46	—
O_2 saturation	82 per cent	82 per cent	—
O_2 capacity, vol. per cent	17.47	16.89	—
A-V O_2 difference, vol. per cent	9.40	5.84	—
Cardiac output, L/Min			
(breathing oxygen)	4.12	4.60	—
Maximal pulse pressure			
Right auricle	13	25	—
Right ventricle	78	68	—

of 350 ml. of blood, was then able to breathe easily while lying in the supine position, and rejected further supplemental oxygen therapy. The observed electrocardiographic K constant ($QT/\sqrt{\text{cycle length}}$) progressively shortened from 0.432 to 0.342 during the same period of time; inversion of T waves in lead II and occasional ventricular premature beats

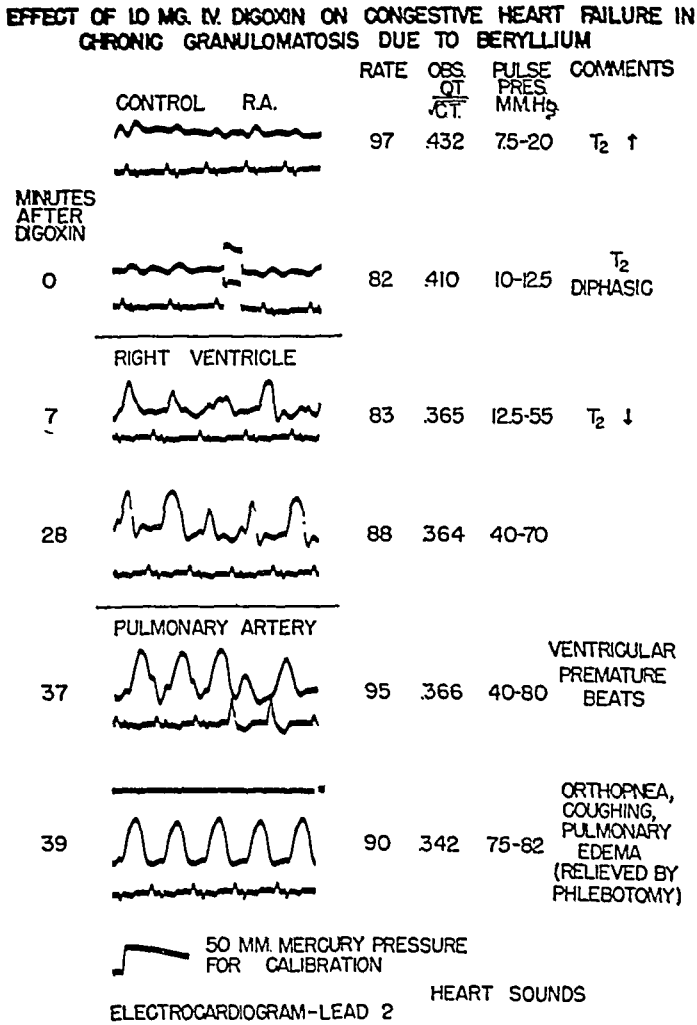


FIG. 6. Effect of digoxin on manifestations of congestive heart failure in patient J. J.; 1.0 mg. digoxin was administered intravenously during the second cardiac catheterrization. (Note irregular right ventricular pressure pulse patterns which were present prior to the administration of the digoxin, as well as typical electrocardiographic changes related to the digoxin).

further indicated the effects of digoxin on the myocardium. The cardiac output increased from 4.6 to 7.8 liters per minute. Diuresis was instituted promptly by the digoxin, and during the first twenty-four hours the basal weight declined by 1.75 Kg. The venous pressure was reduced to 11.0 cm. of water, and the arm-to-lung circulation time shortened to 10 seconds. Patient was thereafter continued on maintenance doses of digitoxin orally. An exercise tolerance test was repeated, and the patient was able to walk on the level the full 10 minute period at 1.7 mph and experienced much less dyspnea. The exercise systemic

pulse pressure was increased to the precongestive-failure level, and the ventilatory equivalent for oxygen was restored to its former value.

These data clearly show how long-standing beryllium granulomatosis can be complicated by cor pulmonale and congestive heart failure. The response to rapid digitalization was quite unusual because of the resistance to pulmonary blood flow which precipitated pulmonary edema requiring a phlebotomy to obtain relief. Such a patient needed continuous digitalis therapy to combat the heart failure.

The extraordinary disability that can result from overwhelming anoxemia is revealed by the sixth patient in this series:

W. R., a 35 year old white married male, was employed in the fluorescent light industry as an engineer. In March, 1945, he first noted exertional dyspnea which progressed in severity. Later he felt irritable, unstable, had anorexia, and during the summer lost 40 pounds in weight. Abnormal markings were found in the chest roentgenogram. Because of orthopnea and paroxysmal coughing, he was given penicillin and streptomycin by inhalation therapy in July, 1947 without striking benefit. Clubbing of the fingers had been present since 1946, and occasionally he noted slight dependent edema. He had had frequent passages of kidney stones for two years. For the six months prior to admission it had been necessary for him to use oxygen by an oral tube continuously to minimize dyspnea. On examination in May, 1948, he appeared to be a severely, chronically ill, white male with extensive wasting from malnutrition. He exhibited respiratory distress even at rest, and this was accentuated by any cause for anxiety. He showed variable tachypnea, dyspnea, orthopnea, as well as very marked clubbing and appreciable cyanosis even while breathing oxygen by a tube in his mouth. With persuasion the maximum possible time that he could tolerate breathing room air was about 20 minutes early in the course of observation, but later this time was less than a minute (figure 7). The chest was somewhat enlarged and the accessory muscles of respiration were used prominently. Breath sounds were of emphysematous character with wheezing and basilar rales bilaterally. The heart was not enlarged, the rhythm was regular, and the second pulmonic sound was accentuated. A faint systolic murmur as well as an early decrescendo diastolic murmur was audible along the left sternal border. Roentgenographic examination of the chest showed diffuse granular and confluent shadows bilaterally but more marked on the right side (figure 1f). The hematocrit was reduced to 37.5 per cent and the urine contained considerable albumin (3 plus reaction), and both red blood cells and white blood cells. The patient frequently passed kidney stones, and spectroscopic examination revealed beryllium within the stones.[7] The blood carbon dioxide combining power was increased to 77 vol. per cent and the chloride was depressed to 88 m.eq. per liter. Two-plus cephalin flocculation and thymol turbidity reactions indicated abnormal hepatic function. Bromsulpthalein retention was 40 per cent (repeated, 20 per cent) at the end of forty-five minutes. The observed blood volume was 5,600 ml., equal to the value predicted from height, but the plasma albumin concentration by electrophoresis was diminished to 2.7 Gm. per 100 cc., and all globulin concentrations except alpha-1 globulin were abnormal. The maximum breathing capacity was greatly depressed to 10.2 liters per square meter, or 15 per cent of the predicted value (table 2). The lung volumes showed marked reduction in total and vital capacities to 40 per cent and 53 per cent of values predicted (figure 2, table 2). The alveolar pressure-velocity relationships showed that less than the average pressure was needed to obtain moderate flow of air in the tracheobronchial tree (table 2). The electrocardiogram showed right heart strain with right axis deviation and inversion of T waves in leads II and III. Despite conspicuous distension of the peripheral veins, the venous pressure was repeatedly found to be between minus 0.5 and 2.0 cm. of water. The circulation times were normal. Venous catheterization of the right heart revealed increased pressure in auricle, ventricle, and

pulmonary artery (table 3). The cardiac output by the direct Fick principle with the patient breathing oxygen was increased to 5.92 liters per minute. Arterial oxygen saturation was 89 per cent with a pO_2 of 95 mm. The pCO_2 was increased to 56 mm. Breathing room air lowered the pO_2 to 46.2 mm. within a minute. The importance of oxygen therapy to this patient was further revealed by the rapid drop in arterial oxygen saturation based upon oximeter readings at 5 second intervals (figure 7). No exercise studies could be made on this patient.

This last patient, W. R., exhibited contracted lungs with relative emphysema, marked arterial anoxemia and cor pulmonale. In addition he suffered from mal-

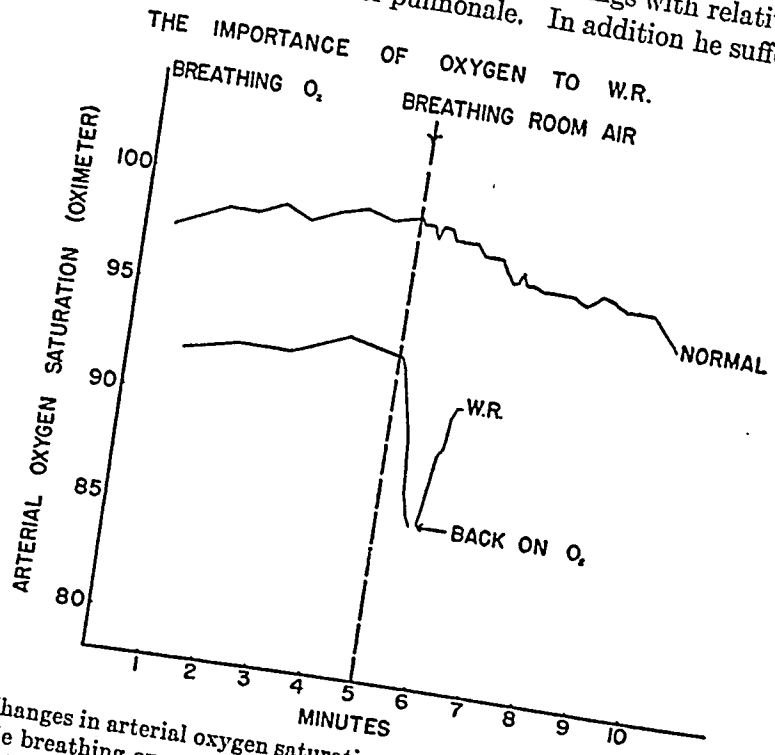


FIG. 7. Changes in arterial oxygen saturation on patient W. R. determined by oximeter readings while breathing oxygen and room air. (Note prompt decline in saturation in 35 seconds at which point the patient demanded restoration of oxygen.)

nutrition, wasting, hepatic, and renal disease. He required continuous oxygen therapy, and had moderate anemia rather than any compensatory polycythemia. The hematuria from renal lithiasis was partly responsible for the anemia. Yet there was no evidence of congestive heart failure, and in fact the venous pressure was subnormal. With deep inspiration the right ventricular diastolic pressure was observed to fall as low as minus 20 mm. of mercury. Because of fear of inducing a fatal pneumothorax, no attempt was made to measure the intrapleural pressure directly which must have been extraordinarily low during inspiration. In order to ascertain if some of the findings might be affected by cytochrome C therapy, the following observations were made (table 6):

The initial effects of cytochrome C observed were a 32 per cent reduction in oxygen consumption but no significant change in either arterial oxygen saturation or tension. The

TABLE 6

*Effect of cytochrome C on patient W. R.**

	BEFORE	AFTER
Breath-holding time, seconds	5	20
Minute ventilation, liters/M^2/minute	5.98	6.44
Ventilatory equivalent for oxygen, liters/minute	2.36	3.73
Oxygen consumption, ml/M^2/minute	253	171
Arterial Blood		
Carbon dioxide tension, mm	75	—
Oxygen tension, mm	67	76
Carbon dioxide, vols. per cent	79.6	76.9
Oxygen content, vols. per cent	12.6	12.6
Oxygen capacity, vols. per cent	14.5	14.7
Oxygen saturation, per cent	87	86
Mixed Venous Blood		
Carbon dioxide tension, mm	84	85
Oxygen tension, mm	35	38
Carbon dioxide, vols. per cent	80.5	78.8
Oxygen content, vols. per cent	8.65	8.85
Oxygen saturation, per cent	59	60
A-V carbon dioxide difference, vols. per cent	0.88	1.86
A-V oxygen difference, vols. per cent	3.93	3.75
Cardiac output liters/minute	6.42	4.56
Heart rate	98	118
Stroke volume, average ml	65	39
Electrocardiogram	$T_{1,3}$—flat	T_1—elevated, T_3—depressed
K constant $\frac{QT}{\sqrt{CL}}$	0.425	0.403
Pneumocardiogram, Chest		
Beginning of right ventricular ejection, sec	0.07	0.05
Beginning of left ventricular ejection	0.13	0.12
Mean right auricular pressure, mm. Hg.**	+2.8	−0.74
Mean right ventricular pressure, mm. Hg	23	24.6
Right ventricular pulse pressure, mm. Hg	66	84

* 80 mg. were administered through a catheter inserted into the right heart. Observations were made immediately before and one-half hour after catheterization while patient was breathing oxygen.

** Zero reference point 5 cm. below level of sternum.

cardiac output declined 29 per cent, with a rise in heart rate and a fall in stroke volume. The sytemic blood pressure rose from 102/72 to 126/100 mm. of mercury. The right ventricular pulse pressure rose from 66 to 84 mm. of mercury as the systemic pulse pressure fell

from 30 to 26 mm. of mercury. The electrocardiogram showed changes in T waves in the limb leads, and a reduction in the K constant. The beginning of ventricular systolic ejection, as indicated by simultaneous electrocardiogram and chest pneumocardiogram, shortened from 0.07 to 0.05 sec. after the beginning of the QRS complex for the right ventricle, and from 0.13 to 0.12 sec. for the left ventricle. The A-V oxygen difference diminished slightly from 3.93 to 3.75 vol. per cent. Maintenance doses of 50 to 75 mg. of cytochrome C were given intravenously daily for five days and, except for doubtful reduction in oxygen consumption, no effects were observed. Symptomatically the patient was unable to describe any specific change.

Based on inconclusive observations in a single patient it is impossible to assess any specific benefit to cytochrome C therapy in this group of patients.

Determination of Alveolar-arterial Oxygen Gradient

Inasmuch as the impaired diffusion of oxygen from the alveolar spaces to the pulmonary capillaries is held to be the keystone to the pathological physiology of beryllium granulomatosis (8), the results obtained in 5 of these patients are graphically portrayed in relation to ventilation, hemoglobin concentration and arterial oxygen saturation (figure 4). In comparison with normal average ventilation volumes at rest, these patients exhibited variable degrees of compensatory hyperventilation ranging from 40 to 240 per cent. The latter value is that for patient W. R., but it is important to note that at the time of this observation he was unable to tolerate breathing room air for more than five minutes. The resting ventilation was still more than 50 per cent above normal even while he was breathing supplemental oxygen. The alveolar pO_2 values recorded in figure 4 are those calculated from Riley's formula:

$$\text{Estimated alv. } pO_2 = (\text{tracheal } pO_2)\,\frac{\text{Exp. } N_2 \text{ per cent}}{\text{Insp. } N_2 \text{ per cent}} - \frac{\text{Art. } pCO_2}{\text{exp. R. Q.}}$$

In every instance but that of patient M. H. C., the observed mid-capacity pO_2 (by means of continuous sampling during breathing with a mask) was appreciably higher than the calculated alveolar value (table 7). The calculated alveolar-arterial pO_2 gradient (calc. alv. pO_2 minus observed arterial pO_2) was significantly increased in all patients, ranging from 24 mm. in patient M. E. T. to 50 mm. in patient W. R. It should be noted that the lowest gradient was found in patient M. E. T. who had the highest level of hemoglobin, 14.8 Gm. per 100 cc. None of these 5 patients exhibited a compensatory polycythemia, in fact 2 or 3 showed minor degrees of anemia. This, of course, contributed to the hypoxemia as well as obscuring frank evidence of visible cyanosis. Thus in these 5 patients, overbreathing was the sole compensatory mechanism for hypoxemia due to increasing the alveolar-arterial oxygen gradient. Dividing the observed oxygen consumption by the alveolar-arterial oxygen gradient, based upon the calculated value for alveolar pO_2, yielded diffusion constants for oxygen that were far below normal values (table 7) (22).

DISCUSSION

The causes of dyspnea found in these patients with pulmonary granulomatosis are essentially similar to those observed by Wright and his associates on a simi-

lar group of patients. Overbreathing, present at rest, and accentuated by exertion, is the result of an increased alveolar-arterial oxygen gradient, hypoxemia, and contracted breathing capacity and total lung volume (particularly the complemental air partition). The latter change may increase the proportion of residual air, but the observed volume was not greatly increased in any of these patients. Thus the pulmonary insufficiency more nearly resembles that seen in patients with pulmonary fibrosis than in those in whom emphysema is the primary defect. Pathological examination of fatal cases has shown the presence of diffuse emphysematous alterations in lung structure intermingled with granulomatous infiltrations. Functional studies of these patients do not, of themselves, offer data that serve to differentiate granulomatosis from other pulmonary diseases of comparable severity. Such studies are of value in delineating the differences between patients of the same group and, when analyzed according to pulmonary efficiency in relation to work performed (figure 3), the sequence of

TABLE 7

*Resting ventilation, alveolar pO_2, alveolar-arterial oxygen gradient, and diffusion constant for oxygen**

PATIENT	RESTING VENTILATION	OXYGEN CONSUMPTION	CALCULATED ALVEOLAR pO_2	OBSERVED ARTERIAL		ALVEOLAR-ARTERIAL OXYGEN GRADIENT	DIFFUSION CONSTANT FOR O_2	VENTILATORY EQUIVALENT FOR O_2
				pO_2	pCO_2			
	L/M²/min.	*ml/M²/m*	*mm.*	*mm.*	*mm.*	*mm.*		
M. C.	4.72	140	111	74	47	37	2.94	3.37
M. E. T.	4.21	162	96	72	36	24	3.60	2.60
M. H. C.	6.80	168	120	78	25	42	4.20	4.03
J. J.	4.24	141	98	49	41	49	2.04	3.03
W. R.	10.21	161	96	46	48**	50	1.97	6.38

* All measurements made while breathing room air.

** pO_2 96 and pCO_2 66 mm. while breathing oxygen.

complications is more readily understood. The complications of pulmonary granulomatosis evolve as compensating mechanisms for hypoxemia and increased pulmonary resistance. The earliest response to hypoxemia is hyperventilation, whereas usually polycythemia is a response to long-standing hypoxemia. Although occasional secondary polycythemia (8) has been observed in patients with granulomatosis, only one (patient A. B.) of those studied here exhibited this phenomenon. The infrequency of this response is provocative and in only patient W. R. could it have been obscured by chronic blood loss (from renal lithiasis). It is possible that dietary deficiencies, inadequate absorption of constituents of hemoglobin, or impaired hemoglobin metabolism related to the generalized beryllium toxicity, may have been responsible. Of particular interest in this regard are the observations of other investigators who have noted the absence of polycythemia in patients with severe chronic anoxemia. Studies on 20 patients with pulmonary tuberculosis treated by thoracoplasty failed to show any evidence of polycythemia despite arterial anoxemia (23). Investigations of the

influence of anoxemia on the hemopoietic activity in healthy and diseased males at high altitudes have shown limitations to the hematologic response to the anoxic stimulus. Extreme anoxia results in a decrease in the polycythemic response which may be attributed to an interference with the formation of hemoglobin (24). Cor pulmonale develops after the onset of pulmonary hypertension. The earliest evidence of this is increased right ventricular pressure with exertion; the late manifestations are right heart strain and congestive heart failure. Data obtained on patient J. J. indicate that retention of water and electrolytes are important contributory factors to the appearance of failure. In addition, the too rapid treatment of congestive failure with intravenous digoxin can be hazardous when the pulmonary vascular bed is unable to accomodate an increased right heart output. Under these circumstances pulmonary edema was acutely precipitated, and just as quickly relieved by phlebotomy. Excessive exertion, such as grade walking, can cause acute transient myocardial failure as manifested by more severe dyspnea and markedly increased oxygen debt in patient M. C. Digitalis therapy appears to be of value only in the presence of congestion, for no greater exercise tolerance was exhibited by its use in patients without congestion. Orthopnea, however, is an expression of myocardial failure in either instance.

Many of the patients with pulmonary granulomatosis are distressed by daily fluctuations in intensity of dyspnea. In part these variations can be related to psychological adjustments to an occupational disease and the effects on their daily activities. As expected, anything that can induce anxiety in these patients aggravates their dyspnea. Finally more profound alterations in water and electrolyte metabolism, especially in relation to the menstrual cycle, may alter awareness of dyspnea. An example is the increased blood volume during the premenstrual phase associated with lessened vital and total pulmonary capacity and greater exertional dyspnea, presumably because of pooling of more blood within the lungs in patient M. E. T. In the same patient the ingestion of ammonium chloride during the menses resulted in weight loss, striking reduction in residual air, and decrease in arterial pCO_2 together with marked accentuation of dizziness. Despite these symptomatic variations, the ventilatory equivalent for oxygen, which expresses pulmonary efficiency, was the least variable datum in a series of multiple exercise tolerance tests performed on 2 patients, M. E. T. and M. H. C.

The secondary effects are those of malnutrition, liver, and renal disease. Appropriate metabolic studies will be the subject of a separate report by Dr. Christine Waterhouse. From preliminary observations it is not yet clear whether hypoxemia alone, or in combination with beryllium poisoning of enzyme systems, can account for these effects. Except for the necessity of maintaining patient W. R. against overwhelming anoxemia, the observed data appear to afford no evidence that any specific benefits were obtained from either oxygen or cytochrome therapy in these patients.

SUMMARY

The resting and exercise measurements of pulmonary and cardiovascular functions in 6 patients with chronic pulmonary granulomatosis are presented in rela-

tion to the evolution of the functional impairment and its complications. The results of these studies are essentially in accord with those observed by other investigators of a similar group of patients. The main deviation from normal is the hyperventilation, especially during work, compensating for hypoxemia. Diminished arterial oxygen saturation is a function of abnormally increased alveolar-arterial gradient for oxygen and contracted lung volume, particularly complementary air. One of these patients exhibited polycythemia as a compensatory mechanism. Various stages of cor pulmonale, including congestive heart failure due to increased pulmonary resistance, were observed. An unusual response to intravenous digoxin and the lack of benefit from cytochrome C therapy are recorded. Variations in response to exercise and similarities to functional impairment of other chronic pulmonary disease are discussed.

Addendum

Two more patients have been studied since the preparation of this report. Each one has presented findings similar to the others, but neither has been as far advanced as either J. J. or W. R.

SUMARIO

Observáciones Relativas a las Causas de la Dispnea en la Granulomatosis Pulmonar Crónica de los Trabajadores en Berilio

Las mediciones de las funciones pulmonar y cardiovascular en descanso y en ejercicio en 6 enfermos con granulomatosis pulmonar crónica se presentan en relacion con la evolución de la disfunción y sus complicaciones. El resultado de estos estudios concuerda esencialmente con lo observado por otros investigadores en un grupo semejante. La principal desviación de lo normal consiste en la hiperventilación, sobre todo durante el trabajo, que compensa la hipoxemia. La hiposaturación arterial para el oxígeno es efecto de la anormal hipergradiente alveoloarterial para el oxígeno y del disminuido volumen pulmonar, en particular para el aire complementario. Uno de los enfermos manifestó policitemia como mecanismo compensador. Observáronse varias etapas del "cor pulmonale," incluso insuficiencia cardíaca congestiva debida a hiperresistencia pulmonar. Menciónanse una inusitada respuesta a la digoxina por vía venosa y la ineficacia de la citocromo-C-terapia. Discútense las varias reacciones al ejercicio y las semejanzas a la disfunción ocasionada por otras neumopatías crónicas.

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THE ROLE OF PULMONARY CAVITATION IN THE DEVELOPMENT OF BACTERIAL RESISTANCE TO STREPTOMYCIN[1, 2]

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INTRODUCTION

The use of antibiotics in combating infection is frequently limited by the capacity of microorganisms to develop resistant strains against which the drugs are no longer effective. This is particularly true in the case of a chronic disease such as tuberculosis and the antibiotic streptomycin which, though an excellent bacteriostatic agent, has little bactericidal effect. In the earliest reported *in vitro* studies, as well as the first attempts to use streptomycin in the treatment of human tuberculosis (1, 2), the development of highly resistant mycobacterial strains was noted. Though much work has been done on this subject during the past two years, there is still no clear cut explanation of the factors involved in the development of resistant strains of *M. tuberculosis*. It is evident that adequate knowledge of these factors would greatly increase the field of usefulness of streptomycin.

That the duration of therapy with streptomycin is one factor in causing the development of resistant bacilli has been shown in experimental animals. Steenken (3), after treating tuberculous guinea pigs with streptomycin for 125 days, recovered only nonresistant tubercle bacilli from the animals. Feldman, Karlson, and Hinshaw (4) obtained similar results when infected guinea pigs were treated for 60 days, but found that, if treatment was continued for 206 days or longer, tubercle bacilli resistant to more than 2,000 γ of streptomycin per ml. of media could be cultured from the spleen of each animal. Also the data from various clinics suggest the important role played by duration of therapy. A report from the Veterans Administration Hospitals (5) indicates that after 60 to 90 days of treatment bacilli resistant to 10 γ or more of streptomycin have developed in 64 per cent of the patients; after 120 days of therapy more than 78 per cent of the patients had developed resistant strains. The work of Youmans and Karlson (6) and that of Bernstein, D'Esopo and Steenken (7) similarly indicate the dangers with regard to bacillary changes inherent in long courses of streptomycin.

While the importance of long continued streptomycin therapy in causing resistant bacteria to develop is recognized, evidently other factors are of greater importance in some cases. For example, Wolinsky, Reginster and Steenken (8), in reporting on 47 patients treated with streptomycin, obtained one culture

[1] From the William H. Maybury Sanatorium (Detroit Municipal Tuberculosis Sanatorium), Northville, Michigan.

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resistant to 1,000 γ of streptomycin per ml. of media after only 29 days of therapy, while cultures from 11 of the patients remained sensitive to 1.0 γ of streptomycin after 12 weeks of therapy. Howlett and O'Connor (9) observed that in 26 patients with disseminated nodular lesions who received streptomycin therapy the only one who eventually produced tubercle bacilli resistant to 10 γ of streptomycin was a patient who also had a persistent cavity. In this particular patient the bacilli eventually became resistant to 1,000 γ of streptomycin.

Prior to the publication of the last mentioned article, the casual observation was made at the Maybury Sanatorium that there seems to be a close correlation between the presence of cavity and the development of resistant tubercle bacilli. To test this observation more carefully, an analysis has been made of the data obtained from 155 streptomycin-treated patients who have had posttreatment resistance studies performed on tubercle bacilli isolated from their sputum or gastric contents.

MATERIALS AND METHODS

The 155 patients included in the present study represent approximately one-half of the adult patients with pulmonary tuberculosis who were treated with streptomycin at the Wm. H. Maybury Sanatorium during the interval from November, 1946 to August, 1948. The only patients excluded from the study are those on whom no posttreatment determinations of bacterial sensitivity to streptomycin are available. For most of the sensitivity determinations we are indebted to Dr. Cora R. Owen[2] of the University of Michigan and to Dr. Guy P. Youmans[2] of Northwestern University.

During the twenty month period of the study, six significantly different streptomycin dosage schedules were employed; 2 Gm. per day given in divided doses of 0.4 Gm. five times daily for 90 days; 1.0 Gm. per day given in divided doses of 0.5 Gm. twice daily to a total of 100 Gm.; 1.0 Gm. daily given as a single dose for 42 days; 0.5 Gm. daily given as a single dose for 84 days; a dose varying from 0.4 to 0.7 Gm., depending upon the patient's weight and given twice daily for 84 days. In one small series of patients streptomycin was given during alternate weeks according to the schedule last mentioned.

Chest roentgenograms were taken at the beginning of therapy and thereafter at intervals of four weeks in most cases; in the case of patients whose therapy was terminated at the end of six weeks the roentgenogram was taken at that time.

In preparing the correlation tables only a single bacterial sensitivity determination has been used for each patient except in 11 instances. In these 11 cases, 2 sensitivity determinations, made on bacteria isolated from a single patient at intervals longer than four weeks, have been used. Thus 166 sensitivity determinations, made on bacteria isolated from 155 patients, are included in the study. In addition to these 166 determinations made during the course of streptomycin therapy, or after its completion, 7 pretreatment determinations were done. All 7 of these cultures failed to grow in media containing one γ of streptomycin per ml.

Data regarding the presence or absence of cavitation were obtained from the interpretation of the roentgenologist as recorded in each patient's chart. If the interpretation was equivocal on this point, the original roentgenogram was consulted. The pertinent roentgenogram was the one taken at the same time, or near the time, that tubercle bacilli were isolated from the patient's sputum for the sensitivity determination. Since most of the sensitivity tests were made at the termination of therapy, this means that in most cases the patient's roentgenogram taken at the *conclusion* of his course of streptomycin determined whether or not he should be classified as having a cavity.

RESULTS OF STUDY

The impossibility of classifying all cases as definitely showing or not showing cavitation soon became apparent. As mentioned above, the original roentgeno-

grams were consulted whenever the recorded reading of the roentgenologist was found equivocal. Even with the additional information obtained from subsequent roentgenograms, however, it was frequently impossible to say whether or not at a given time definite cavitation was present. Thus, instead of separating the cases into two groups, those with and those without cavities, it was found necessary to add a third group of cases in which the findings with regard to the presence or absence of cavities remained equivocal. In the first group, those showing definite cavitation at the time the sensitivity determination was made, there are 88 cases. Thirty-seven cases are found in the "equivocal" group, while 41 cases are included in the noncavity group. In 11 instances, patients who showed definite cavitation at the beginning of therapy had completely closed the cavity by the time the sensitivity determination was made. In 21 instances, patients with cavitation at the beginning of therapy were finally placed in the "equivocal" group. Changes in the reverse direction occurred only four times

TABLE 1

Data from cases showing cavitation at the end of streptomycin therapy

RESISTANCE TO STREPTOMYCIN	DURATION OF THERAPY IN DAYS			TOTALS		
	28 to 41	42 to 83	84 to 160	Number	Per cent	
	Number of cultures					
γ per ml.						
<1	0	4	0	4	4.5	
1–10	5	5	0	10	11.4	15.9
10–100	2	12	10	24	27.3	
100–1,000	2	7	13	22	25.0	
>1,000	3	17	8	28	31.8	84.1
<1–>1,000	12	45	31	88	100.0	100.0

while patients were taking streptomycin and in four instances patients who had no cavity at the beginning of therapy had roentgenograms which were considered "equivocal" at the time the sensitivity determination was made.

With 6 exceptions, every patient who had definite cavitation at the beginning of therapy had sputum which was positive for acid-fast bacilli on direct smear. The 6 exceptions were "positive" only on culture of the sputum.

The data for the three groups of patients, classified according to duration of therapy and the level of streptomycin resistance of the mycobacteria isolated from the sputum, are shown in tables 1, 2, and 3.

Following established custom (5), it is assumed that tubercle bacilli able to grow in media containing 10 γ of streptomycin per ml. are *resistant* as far as effective clinical therapy is concerned. Using this definition of resistant microorganisms, it will be seen from table 1 that of the patients showing cavitation the great majority (84.1 per cent) produced resistant tubercle bacilli. Among the cultures obtained 28 to 41 days after the beginning of therapy, 7 of 12, or 58.3

per cent, were resistant. In the 42 to 83 day group, 36, or 80 per cent of resistant cultures, were found and in the 84 day group *all* of the cultures were resistant.

It will be noted that *no* cultures of tubercle bacilli *sensitive* to one γ or less of streptomycin were found in the 28 to 41 day group, while four cultures of this type were found among patients who had had therapy for 42 to 83 days. This obvious aberration can be explained on the basis of the composition of the various

TABLE 2

Data from cases in which the evidence of cavitation remained equivocal

RESISTANCE TO STREPTOMYCIN	DURATION OF THERAPY IN DAYS			TOTALS		
	28 to 41	42 to 83	84 to 160	Number	Per cent	
	Number of cultures					
γ per ml.						
<1	1	7	0	8	21.6	
1–10	2	20	3	25	67.6	89.2
10–100	0	1	0	1	2.7	
100–1,000	0	1	0	1	2.7	
>1,000	0	1	1	2	5.4	10.8
<1–>1,000	3	30	4	37	100.0	100.0

TABLE 3

Data from cases showing no cavitation at the end of therapy

RESISTANCE TO STREPTOMYCIN	DURATION OF THERAPY IN DAYS			TOTALS		
	28 to 41	42 to 83	84 to 160	Number	Per cent	
	Number of cultures					
γ per ml.						
<1	3	10	6	19	46.3	
1–10	2	13	5	20	48.8	95.1
10–100	0	1	1	2	4.9	
100–1,000	0	0	0	0	0	
>1,000	0	0	0	0	0	4.9
<1–>1,000	5	24	12	41	100.0	100.0

groups as regards dosage schedule. In the first group, the majority of the 12 patients had been on the 0.5 Gm. schedule, *i.e.*, they had received 0.5 Gm. of streptomycin daily in a single dose. The 42 to 83 day group included only one patient who had been on the 0.5 Gm. schedule and 34 who had received 1.0 Gm. daily in a single dose. The remaining 10 patients had received either 0.5 Gm. twice daily or 2.0 Gm. daily divided into five doses. In the light of these different dosage schedules, the complete absence of any cultures in the 28 to 41 day group sensitive to one γ or less of streptomycin suggests that the daily dose of 0.5 Gm.

of streptomycin *for patients having pulmonary cavities*' merely stimulates the population of tubercle bacilli present in the cavities to an early development of resistance.

An observation similar to the above is mentioned by Steenken (10). After 46 to 90 days of therapy, he found that a greater proportion of patients receiving 0.5 Gm. of streptomycin per day had produced resistant tubercle bacilli than was the case for those patients receiving either 1.0 Gm. or 1.8 Gm. daily. Also pertinent to this question is the report of Wolinsky and Steenken (11) that tubercle bacilli begin growing *in vitro* with little or no delay in a culture medium

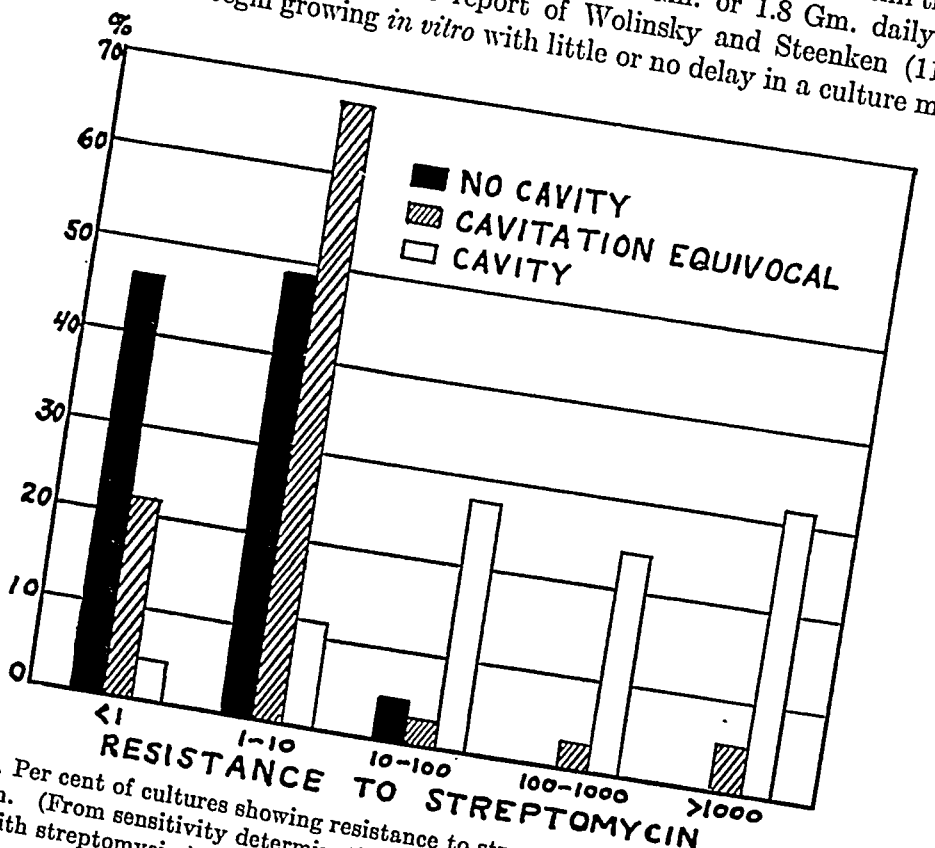

FIG. 1. Per cent of cultures showing resistance to streptomycin as related to persistent cavitation. (From sensitivity determinations on 166 cultures obtained from 155 patients treated with streptomycin.)

containing 0.1 γ of streptomycin per ml., while there is no growth for more than two weeks in the same medium containing 0.2 γ of streptomycin per ml.

To be noted in table 1 is the fact that in 3 instances tubercle bacilli resistant to more than 1,000 γ of streptomycin were found after only 28 to 41 days of therapy. All 3 of these patients had received the minimal dose of 0.5 Gm. of streptomycin daily, one of them for only 28 days, the other 2 for 34 days. All 3 had large cavities measuring from 5 to 10 cm. in diameter.

Table 2 shows that 10.8 per cent of the patients in whom the finding with regard to cavitation was equivocal developed resistant strains of tubercle bacilli. In 3 instances the original area of excavation became opaque to the roentgen

rays and was interpreted as a "filled" cavity. The bacilli from 2 of these patients were sensitive to 10 γ of streptomycin; bacilli from the third fell in the 10 to 100 γ per cc. resistance group.

In table 3 only 2 patients (4.9 per cent of the total) are found to have developed resistant strains. One of these 2 patients had tuberculous pericarditis as well as disseminated pulmonary disease; the other had received streptomycin for 115 days, except for one intermission of two weeks, when the resistant culture was obtained.

Of further interest in table 3 is the fact that 6 patients, after treatment with streptomycin for 84 days or longer, still harbored tubercle bacilli which were sensitive to 1 γ of streptomycin. Five of the 6 began their treatment with recently formed cavities which closed during the course of streptomycin therapy; the sixth did not show a cavity at any time. Most of these particular patients have been mentioned in a previous article (12). Subsequent follow-up shows that all 6 have become "sputum-negative"; their roentgenograms have shown progressive clearing and their prognosis is excellent.

The data from the final columns of tables 1, 2 and 3 are presented graphically in figure 1. This chart emphasizes the fact that in the presence of persistent cavitation the development of resistant strains of tubercle bacilli is a significant matter. In the absence of cavitation at the end of treatment, resistant strains rarely develop. In those cases in which cavitation is equivocal there is still no significant development of resistant strains.

DISCUSSION

The data presented above illustrate the marked variation observed in clinical cases of tuberculosis with regard to the development of resistant strains of tubercle bacilli. In one group of cases the bacilli remained sensitive to one γ of streptomycin even after 84 days of therapy; in another group resistance to more than 1,000 γ of streptomycin developed after only four to six weeks of therapy. What is the reason for this variation? Is the development of resistant tubercle bacilli, as suggested by Yegian and Vanderlinde (13), merely a consequence of the ". . . selection of normally occurring hereditary variants that are present in the original cell population"? If this were true, then the bacilli of a patient in the first group would remain sensitive because that patient's "original cell population" did not contain resistant cells, whereas a patient in the second group would be unlucky because amongst his original quota of bacilli there happened to be highly resistant cells. In the light of the data which have been presented, this explanation seems unlikely. There is too close a correlation between persistent cavitation and the development of resistant bacilli to be explainable on the basis of mere chance variation in the original cell population.

One obvious reason for the importance of the cavity in determining whether or not resistant bacilli will appear is the well known fact that tubercle bacilli grow much more readily and more rapidly in this location where they have been "exteriorized" as far as the ordinary defense mechanisms of the body are concerned (14). Rapid multiplication is generally accepted as important in the

development of resistant microorganisms (2, 15). Another important characteristic of tuberculous cavities, particularly the older, more chronic variety, is that their relatively avascular walls (16) militate against streptomycin reaching the interior of the cavity in effective concentration. In one experimental study (17) the streptomycin content of sputum from the cavities of patients under therapy with 1.8 to 2 Gm. of streptomycin daily was found to vary from a minimum of 1 γ per ml. to a maximum of 18 γ per ml.

Finally, within the tuberculous cavity the tubercle bacillus is frequently able to grow in a fluid or semifluid menstruum instead of on the surface of the cells, and this may be of considerable importance in bringing about the rapid evolution of highly resistant strains. Steenken has stated (18) that strains of tubercle bacilli resistant to streptomycin can be produced much more rapidly when the microorganisms are grown in the depths of a liquid medium such as that of Dubos (19) than when they are grown as a pellicle on the surface of a medium. Ballon and Guernon (20, 21) and Follis (22) have shown that sulfanilamide, incorporated in a solid medium, completely inhibits the growth of tubercle bacilli while sulfanilamide in the same concentration in a liquid medium has no inhibitory effect. These observations suggest that strains of tubercle bacilli resistant to sulfanilamide may evolve when the bacteria are grown in liquid but not on solid media containing the drug.

From the data of the present study it is evident that strains of tubercle bacilli highly resistant to streptomycin may be produced very rapidly in some cases while in other cases the increase in resistance on the part of the tubercle bacillus appears to be a gradual process. Demerec (23) has noted a similar behavior in the case of the staphylococcus. When this microorganism is submitted to the action of streptomycin for a single time, some strains are produced which are almost completely resistant while other strains are only slightly more resistant than the parent strain. Demerec explained this erratic sort of increase in resistance to streptomycin on the basis of mutations in genes varying greatly in their potency, a mutation in a highly potent gene being responsible for a high degree of resistance, a mutation in a less potent gene for a low degree of resistance. The hypothesis that similar changes may occur in the case of the tubercle bacillus offers an attractive explanation for the observed facts.

Just how rapidly may highly resistant strains of tubercle bacilli appear? Vennesland *et al.* (24) have demonstrated that tubercle bacilli 10,000 times as resistant to streptomycin as the original strain may originate from a stock laboratory strain of H-37Rv after its exposure to dilute streptomycin for 21 days in Dubos medium[3]. With regard to clinical studies, one patient in the present series discharged tubercle bacilli resistant to 1,000 γ of streptomycin after 28 days of therapy. Wolinsky, Reginster and Steenken (8) have reported that a strain similarly resistant appeared after 29 days of therapy, and McDermott (25) noted the appearance of a highly resistant strain after five weeks of therapy. The most specific example of the rapid emergence of resistant tubercle bacilli

[3] This interpretation is different from that of Vennesland *et al.*, who considered that naturally occurring streptomycin-resistant variants had been demonstrated.

has been mentioned by Karlson (26) who made sensitivity tests on tubercle bacilli isolated on the dates mentioned below from the sputum of a patient receiving streptomycin therapy[4].

Date of sputum culture	*γ of streptomycin per ml. of culture medium necessary to inhibit growth*
3-22-46	0.31
3-28-46	0.31
4- 1-46	0.31
4-10-46	0.31
4-17-46	0.31
4-26-46	0.31
5- 8-46	> 1,000

Thus, after remaining sensitive to 0.31 γ of streptomycin for approximately six weeks, the tubercle bacilli isolated from this particular patient only 12 days later were resistant to more than 1,000 γ of streptomycin.

It is evident that the cumulative effect of numerous small step mutations (such as were observed by Demerec), should they occur in the tubercle bacillus, would eventually produce highly resistant bacilli. Against this explanation in the present case is the fact that the growth of the tubercle bacillus is necessarily slow, too slow to allow any large number of generations to occur during the 12 day interval observed by Karlson.

In vitro experimental studies indicate that different growth mechanisms may operate for the tubercle bacillus, depending on whether the microorganism is grown in liquid or on solid media. Kahn (27) observed that tubercle bacilli growing in a liquid medium pass through a developmental cycle which requires 17 days for its completion. Tubercle bacilli growing on solid media do not pass through any such cycle but appear to proliferate by direct fission (28). As long as mycobacteria are growing by direct fission, the mycelium which is formed might be compared to the continuous growth of one plant, and no great variation in the individual cells would be expected. However, when the bacillus passes through a complex cycle of development, there is greater opportunity for the mutation of highly potent genes which, in the present case, could bring about abruptly the appearance of a strain of bacilli highly resistant to streptomycin. Of interest in this regard is the observation of Wolinsky and Steenken (11) that tubercle bacilli exposed in a liquid medium to 0.2 γ of streptomycin per ml. show no evidence of growth for the first *17 days*; after this period growth is rapid. The appearance of the 17 day interval in the above work and in the cycle observed by Kahn may be more than a mere coincidence. Also, the 21 day interval found necessary by Vennesland *et al.* (24) for the production of tubercle bacilli highly resistant to streptomycin is long enough to include the 17 day growth cycle.

Returning to experimental tuberculosis in guinea pigs, Feldman, Karlson, and

[4] In following the development of bacterial resistance in streptomycin-treated patients, Steenken (18) has noted that the cultures isolated from a given patient may change from sensitive to 0.5 γ of streptomycin per ml. to resistant to 1,000 γ per ml. in a period of only seven days.

Hinshaw (4), as already mentioned, found that treatment of the animals with streptomycin for a long period of time (206 days) was necessary before highly resistant strains of tubercle bacilli appeared. In this respect tubercle bacilli in the guinea pig react much as they do in the human patient who has no pulmonary cavity. The reason may be the same in both instances, for cavitation is almost never seen in ordinary inoculation tuberculosis of guinea pigs.

The practical implications of the close correlation observed between persistent cavitation and the development of resistant strains of tubercle bacilli are obvious. If there is no cavitation in cases of minimal tuberculosis, the usual course of streptomycin might be given to such patients with little or no fear that resistant bacilli will develop. If cavitation is present and closure of the cavity occurs during the course of therapy, again there is very little danger of the development of resistant strains of tubercle bacilli. These facts should influence the therapeutic strategy of the physician with regard to each patient; his every effort should be directed toward obtaining cavity closure before completion of the streptomycin therapy. If the cavitation is very extensive or of such a character that it may not close during the course of streptomycin, the drug should supplement collapse therapy rather than precede it.

One other implication has to do with the question of streptomycin-resistant tubercle bacilli in patients who have become "sputum negative." These are the patients who have shown such a good response to streptomycin that it is impossible to recover tubercle bacilli from their sputum at the end of the course of therapy. Fears have frequently been expressed, however, that these patients might be harboring highly resistant tubercle bacilli in some obscure focus in the body. In view of the almost complete absence of resistant bacilli in patients without cavitation, these fears seem unfounded.

Data regarding the development of streptomycin-resistant tubercle bacilli in relation to the various dosage schedules employed in the present series of cases are too fragmentary to permit any observations other than that the dose of 0.5 Gm. of streptomycin per day *in cases showing cavitation* appears to be too small to prevent the rapid emergence of highly resistant strains of bacilli. Looking upon the cavity wall as a semipermeable membrane (relatively impermeable in the case of thick-walled cavities), it is evident that a high streptomycin blood level, if maintained for only a few hours, might be more effective in producing a satisfactory level of streptomycin within the cavity than a lower blood level maintained for a longer period of time. Once a fair concentration of streptomycin has been obtained in the contents of the cavity, this level might be expected to remain fairly constant (even after the blood level has fallen) until the cavity is emptied.

Apparently a single 1.0 Gm. dose of streptomycin produces a satisfactorily high blood level (29). Thus 1.0 Gm. of streptomycin given in a single dose on alternate days might be considerably more effective than 0.5 Gm. of the drug given daily. Such a method of "concentrated therapy" at relatively long intervals might make it possible to avoid the cumulative toxicity of long continued daily dosage with streptomycin while maintaining an effective attack against the tubercle bacillus.

SUMMARY AND CONCLUSIONS

One hundred fifty-five cases of pulmonary tuberculosis treated with streptomycin have been analyzed with regard to the emergence of resistant strains of tubercle bacilli in relation to the presence or absence of cavities. From the analysis the following conclusions seem justified:

(*1*) In the presence of persistent cavitation the emergence of resistant strains of tubercle bacilli is a significant matter.

(*2*) In the absence of cavitation at the end of the course of therapy, resistant strains rarely appear.

(*3*) In those cases in which cavitation is equivocal the emergence of resistant strains of tubercle bacilli occurs with appreciably less regularity than in the presence of definite cavitation.

SUMARIO Y CONCLUSIONES

El Papel de las Cavernas Pulmonares en la Producción de Resistencia Bacteriana a la Estreptomicina

Cinento cincuenta y cinco casos de tuberculosis pulmonar tratados con estreptomicina son analizados con respecto a la emergencia de cepas resistentes de bacilos tuberculosos en relacion con la presencia o ausencia de cavernas. El análisis parece justificar las siguientes conclusiones:

(1) En presencia de cavernas persistentes la emergencia de cepas resistentes de bacilos tuberculosos es significativa.

(2) En ausencia de cavernas rara vez aparecen cepas resistentes al terminar la terapéutica.

(3) En los casos en los que es dudosa la presencia de cavernas, la emergencia de cepas tuberculosas resistentes ocurre con regularidad apreciablemente menor que en presencia de cavernas bien definidas.

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SENSITIVITY OF TUBERCLE BACILLI TO STREPTOMYCIN[1, 2, 3]

The Influence of Various Factors Upon the Emergence of Resistant Strains

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INTRODUCTION

The frequent emergence of streptomycin-resistant strains of tubercle bacilli in tuberculous patients treated with streptomycin is now a well known phenomenon (1 to 25). The great importance of these resistant strains in sharply limiting the effectiveness and clinical usefulness of streptomycin is equally well recognized. An increasing number of studies have been stimulated by an interest in possible methods for reducing the incidence of streptomycin-resistant tubercle bacilli and for circumventing the therapeutic obstacles which they create. Of the many variables which might influence the emergence of resistant strains, interest thus far has been largely concentrated upon: (*1*) variations in the daily dose, in the frequency of administration and in the total duration of streptomycin treatment (the regimen employed); (*2*) therapeutic combinations of streptomycin and other drugs. Up to the present time little attention has been given to possible influences inherent in the patient or in the tuberculous disease being treated.

In a detailed report of the study at the Laurel Heights Sanatorium of streptomycin in pulmonary (and bronchopulmonary) tuberculosis (7) which was submitted in February, 1948, analysis of sensitivity data available at the time resulted in the following comments:

"Among patients on Regimen A or B [1.8 or 2.0 Gm. of streptomycin daily for 120 days] strains requiring 50 γ per cc. or more for inhibition[5] emerged only among those patients with frank caseation or cavitation. . . . It appears from our study that lesions in which caseation and cavitation were prominent were not only least likely to show adequate therapeutic response to streptomycin alone, but were also most likely to produce strains of organisms with significant resistance to the drug."

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[1] From the Laurel Heights State Tuberculosis Sanatorium, Shelton, Connecticut, and the Department of Internal Medicine, Yale University School of Medicine.

[2] This study is part of the Streptomycin-Tuberculosis Research Project of the American Trudeau Society, Medical Section of the National Tuberculosis Association. Streptomycin used in the earlier portion of the study was generously donated to the Society by the Abbott Laboratories, Eli Lilly and Company, Merck and Company, Inc., Charles Pfizer and Company, Inc., E. R. Squibb and Sons, and the Upjohn Company. Subsequently the study was continued as part of the Comparative Regimens Study of the U. S. Public Health Service.

[3] This study was aided by grants from the Division of Research Grants and Fellowships of the National Institute of Health, U. S. Public Health Service, and from the Fluid Research Fund of Yale University School of Medicine.

[4] With the technical assistance of Mr. John Vadney, Mr. Hilary Morris, Miss Eleanora Falco, and Mr. Adolph Maruschak.

[5] Since the tests used in this study contained no dilution between 10 and 50 γ of streptomycin per cc., this could also be stated as "strains uninhibited by 10 γ per cc. or less."

Subsequently, the response of certain types of subacute and chronic pulmonary tuberculosis treated with streptomycin was the subject of a special report (8). It was observed that resistant strains of tubercle bacilli (uninhibited by 10 γ per cc. of streptomycin) occurred in none of a group of patients with disseminated nodular pulmonary tuberculosis unless cavity was also present. There was a similar complete absence of highly resistant tubercle bacilli in a group of patients, termed "grumblers", who were treated for other types of persistently active but indolent chronic noncavitary pulmonary tuberculosis. Experience in these two groups was in marked contrast to the experience in patients with frankly caseous or cavernous disease in whom the emergence of highly resistant strains had occurred with significant frequency, even when streptomycin was administered for only 42 days. Again, therefore, there appeared to be a very significant relationship between the incidence of streptomycin-resistant tubercle bacilli and the type of tuberculosis treated.

Complete sensitivity data are now available on a considerably larger number of patients treated for various types of pulmonary tuberculosis. The principal purpose of the present report is to survey in detail two relationships, (*1*) between the incidence of streptomycin-resistant tubercle bacilli and the treatment regimen employed; (*2*) between the incidence of streptomycin-resistant tubercle bacilli and the type of pulmonary tuberculosis being treated.

ANALYSIS OF SENSITIVITY DATA

Materials and Methods

Specimens of sputum or gastric contents from streptomycin-treated patients were collected before treatment and at regular intervals thereafter. These specimens were cultured on solid media at the Laurel Heights Sanatorium and the tubercle bacilli thus isolated were then tested for sensitivity. Sensitivity tests on the great majority of the patients in this report were performed at the Yale University School of Medicine by three of the present writers who have reported elsewhere (9, 10) details regarding the method employed. Briefly, the bacilli were first subcultured in a modified Dubos liquid medium and the subculture was used for the inoculation of tubes of the same medium which contained the following dilutions of streptomycin: 0 (control), 0.5, 1.0, 5.0, 10, 50, 100, 500, and 1,000 γ per cc. Sensitivity was determined as the lowest concentration of streptomycin in which no growth could be detected after ten to fourteen days of incubation at 37°C. Very recently the sensitivity tests for the Laurel Heights study have been performed, instead, at the Trudeau Laboratory by Steenken (11) and his associates, using solid medium containing 0 (control), 1.0, 10, and 100 γ of streptomycin per cc. Thus far, however, complete sensitivity data from this source have become available on only a very few patients.

For purposes of this analysis (and for consistency with other reports submitted on patients in the Laurel Heights study) the streptomycin treatment regimens which have been explored are designated as follows:

Regimen A: 1.8 Gm. daily for 120 days.

Regimen B: 2.0 Gm. daily for 120 days.

Regimen C: 1.0 Gm. daily for 42 days, later revised to 20 mg. per kg. of body weight daily for 45 days.

Regimen III: 40 mg. per kg. of body weight daily, two consecutive days per week (with no streptomycin on the other days of the week) for a total of 13 weeks.

Regimen IV: 20 mg. per kg. of body weight daily for 90 days.

Because of the close similarity of Regimens A and B, patients on these two regimens are

reported together in one group. Regimens III and IV have been used in only a few patients and complete sensitivity data are not yet available on enough of them to justify their inclusion in the analysis by regimens (table 1). All patients with adequate sensitivity data (whatever the regimen employed) have, however, been included in the analysis by type of disease (table 2).

In 8 patients in table 2, sensitivity data were obtained from the Trudeau Laboratory where the test contained no dilution of streptomycin between 1.0 and 10 γ per cc. Pretreatment specimens in all 8 showed growth in 1.0 γ per cc.; 7 of the 8 showed no growth in 10 γ per cc.; one of the 8 showed two colonies in 10 γ per cc., none in 100 γ per cc. In the remaining patients in table 2 and in all patients in table 1, sensitivity data were obtained from the Yale University School of Medicine. On the basis of the test in liquid medium employed there, pretreatment cultures were regularly sensitive to either 0.5 or 1.0 γ per cc. except in 5 patients. In each of these 5 instances a pretreatment reading of 5.0 γ was obtained, although cultures from all 5 patients also had readings of 0.5 or 1.0 γ on subsequent tests.

Unless otherwise specified, all the data which follow are based on at least one or more cultures obtained at or after the end of treatment (as well as during treatment). When the last positive culture during treatment was still sensitive, but sputum and gastric contents became consistently negative prior to the end of treatment and remained so, the result has been classed as indeterminate. This has been done to avoid finally classifying any patient's tubercle bacilli as highly sensitive or intermediate unless the effect upon sensitivity of the entire course of streptomycin is known. This report is concerned, however, only with the effect of the primary course of streptomycin upon sensitivity. A few patients have subsequently been retreated, but data on the effect of retreatment upon sensitivity are too few for analysis, and such data have been excluded from the tables which follow.

In tables 1 and 2 the strains of *M. tuberculosis* isolated from the patients are classified as follows:

1. Highly resistant, *i.e.*, requiring more than 10 γ per cc. to inhibit growth[6];
2. Intermediate, which is subdivided into (a) cultures inhibited by 10 γ per cc. but not by 5, and (b) cultures inhibited by 5 γ but not by 1.0;
3. Highly sensitive, *i.e.*, cultures inhibited by 0.5 or 1.0 γ per cc.;
4. Indeterminate, *i.e.*, no positive culture could be obtained after the end of treatment but all cultures during treatment were inhibited by 5.0 γ per cc. (1 patient) or less (all other patients in this group).

Analysis by Regimens

Complete sensitivity data are now available on 28 patients treated on Regimens A or B and on 47 patients treated on Regimen C. These data are tabulated in table 1.

In 4 patients on Regimen C, strains resistant to 1,000 γ per cc. eventually emerged in spite of the fact that cultures at the end of treatment were still sensitive to 5.0 γ per cc. or less. In these patients the highly resistant strains first appeared in specimens obtained several weeks after the completion of treatment. Amberson and Stearns (12) and Wolinsky, Reginster and Steenken (13) have also reported cases treated for 42 days in which increasingly resistant strains have been demonstrated after cessation of treatment.

In view of the relatively small number of cases reported, it was thought best not to attempt further subdivision in table 1. It is of interest, however, that

[6] On the basis of the tests employed in this study, cultures which grew in 10 γ per cc. required 50 γ per cc. or more for inhibition. Actually the majority of patients classed as resistant eventually discharged bacilli which were uninhibited by 1,000 γ per cc.

among the 28 patients treated on Regimens A or B, frank caseation or cavity was judged to have been present prior to treatment in 18 (64 per cent) and absent in 10 (36 per cent). Among the 47 patients treated on Regimen C, frank caseation or cavity was judged to have been present prior to treatment in 26 (55 per cent), absent in 21 (45 per cent). There is, therefore, no apparent basis for attributing the failure of Regimen C to reduce the incidence of streptomycin-resistant tubercle bacilli to a higher proportion of unfavorable cases in this group.

Analysis by Types of Pulmonary Tuberculosis

For this analysis, patients treated on various regimens have been divided into two broad categories according to the pulmonary disease present prior to treatment. The division has been based almost entirely on the interpretation of pre-treatment roentgenograms, although this interpretation was made with a knowledge of collateral clinical and laboratory findings. Planigrams were used freely whenever the presence of cavity was considered questionable on examination of the conventional films.

TABLE 1

Correlation of sensitivity data to the treatment regimen employed

SENSITIVITY OF TUBERCLE BACILLI TO STREPTOMYCIN AT OR AFTER END OF TREATMENT	REGIMEN A OR B PATIENTS AND PER CENT		REGIMEN C PATIENTS AND PER CENT	
Resistant (> 10 γ)		8 (29%)		13 (28%)
Intermediate: a) 10 γ	5 (18%)	7 (25%)	2 (4%)	10 (21%)
b) 5 γ	2 (7%)		8 (17%)	
Sensitive (0.5 or 1.0 γ)		7 (25%)		19 (40%)
Indeterminate*		6 (21%)		5 (11%)
Total		28 (100%)		47 (100%)

* Last positive culture during treatment sensitive to 5.0 γ (1 patient) or to 0.5 or 1.0 γ (10 patients) but with cultures consistently negative prior to the end of treatment and subsequently.

On this basis, frank caseation or cavity was judged to have been present prior to treatment in 56 patients, while in 31 patients there were no lesions deemed to be frankly caseous or cavernous. Sensitivity data for the two groups are recorded in table 2.

Of the 56 patients with caseation or cavity recorded in table 2: 18 (32 per cent) were treated on Regimen A or B, 26 (47 per cent) were treated on Regimen C, 4 (7 per cent) were treated on Regimen III, and 8 (14 per cent) were treated on Regimen IV. Of the 31 patients without caseation or cavity recorded in table 2: 10 (32 per cent) were treated on Regimen A or B, and 21 (68 per cent) were treated on Regimen C[7]. In the group with caseation or cavity, therefore, a

[7] In a few patients there were slight deviations in either daily dose or duration from the exact regimens listed, but in no instance was such deviation more than minor. When toxic manifestations of streptomycin caused treatment to be terminated within less than 42 days, the patient has been excluded from this report.

somewhat larger proportion of patients received streptomycin for periods of time longer than 45 days than was the case in the group without caseation or cavity. It is possible that the higher incidence of resistant strains in the caseous-cavernous group may be attributable, in part, to this fact. On further analysis, however, it is apparent that this is not the principal factor involved.

All of the patients on Regimens III and IV were patients with gross cavitation, and most of them were deliberately chosen for this reason in order to explore the relative effect of these regimens upon resistance. In 9 of these patients sensitivity after 45 days of treatment (as well as after the end of treatment) is known, and in 7 of the 9 (including all 4 patients on Regimen III) highly resistant strains were demonstrated to have been present in large numbers by the end of 45 days. When these patients are added to the 13 patients on Regimen C, who produced highly resistant strains, there is a total of 20 patients (36 per cent of the entire group of patients with caseation or cavity) in whom resistant strains are known

TABLE 2

Correlation of sensitivity data to the type of pulmonary tuberculosis present prior to treatment

SENSITIVITY OF TUBERCLE BACILLI TO STREPTOMYCIN AT OR AFTER END OF TREATMENT	CASEATION OR CAVITY PRESENT PATIENTS AND PER CENT	NO CASEATION OR CAVITY PATIENTS AND PER CENT
Resistant (> 10 γ)	33 (59 %)	0
Intermediate: a) 10 γ	4 (7 %)	4 (13 %)
b) 5 γ	11 (20 %)	6 (19 %)
Sensitive (0.5 or 1.0 γ)	9 (16 %)	17 (55 %)
Indeterminate*	3 (5 %)	8 (26 %)
Total	56 (100 %)	31 (100 %)

* See footnote, Table 1.

to have emerged as the result of *45 days or less* of streptomycin treatment. This figure (as well as the 59 per cent total incidence of resistant strains in the caseous-cavernous group) is in marked contrast to the failure thus far of highly resistant strains to emerge in force in any patient *without* frank caseation or cavity. Evidence in this study that the type of tuberculosis being treated greatly influenced the incidence of streptomycin-resistant tubercle bacilli, regardless of the treatment regimen employed, appears, therefore, to be unequivocal.

DISCUSSION

General Considerations

It is apparent from several reports (11, 13, 14, 15, 16, 22, 24, 25) that the specific sensitivity data obtained in a given study are considerably influenced by variables in the method of testing, such as the culture medium employed, the size of the inoculum, the incubation time before the test is read, et cetera. Especially noteworthy is the fact that tests in a liquid medium yield results which indicate merely the presence or absence in the inoculum of drug-resistant organ-

isms in sufficient numbers to support growth. An unknown proportion of the patient's bacilli may still be far more sensitive than such a reading indicates. These variables lead to reservations in the interpretation of sensitivity data now available, especially when results from one laboratory are compared with those from another in which different methods were employed. For this report, however, all the tests recorded in table 1 and all but a very few of those recorded in table 2 were performed in the same laboratory by the same method. Moreover, the sensitivity level reported for each patient's bacilli is generally based on repeated tests performed both during and after treatment. Thus the influence of technical factors upon the *comparative* data in the groups reported here has been minimized.

It is difficult to establish an absolutely uniform relationship between *in vitro* resistance and *in vivo* resistance. This is not surprising when one considers not only the variables listed in the preceding paragraph but also the variables in the spontaneous behavior of pulmonary tuberculosis which have always made accurate evaluation of a single factor so difficult. Nevertheless, as Walker (17) has said (in summarizing data from the joint streptomycin study of the Army, Navy, and Veterans Administration), there is an accumulating mass of evidence, no one piece decisive in itself, which indicates that the observation of *in vitro* resistance has very real clinical significance. Experience in the Laurel Heights study strongly supports this conclusion. To say the least, streptomycin is very unlikely to affect a patient's clinical course favorably, once resistant strains of tubercle bacilli appear in sufficient numbers to be demonstrated by the tests in liquid media which have been generally employed thus far in the study of this phenomenon.

The exact level at which *in vitro* resistance become clinically significant is far more dubious. On the basis of both theoretical considerations and practical experience (11, 17, 18, 19, 20) there appears to be good reason for classifying as significantly resistant those strains of tubercle bacilli which grow readily in culture media containing 10 γ of streptomycin per cc. and which require 50 γ per cc. or more to inhibit growth. Certainly bacilli which do not grow in concentrations of 1.0 γ per cc. or less must be considered highly sensitive. Patients who discharge bacilli with *in vitro* sensitivity between these two extremes have been tabulated separately in table 2.

The total incidence of streptomycin-resistant strains of tubercle bacilli encountered in this study (28 per cent of all patients included in table 1, 38 per cent of all patients included in table 2) is considerably lower than that reported from certain other sources (11, 17, 20, 21). It is possible that this difference results in part from the variables already discussed. It is undoubtedly a consequence, in part, to a difference in the method of reporting. In this report, the incidence of drug-resistant strains is expressed as a percentage of the *total* number of patients in a given category; in other reports the incidence has usually been expressed as a percentage of the number of positive cultures (or of patients producing positive cultures) at a specified interval of time after the start of treatment. It is believed, however, that a difference in case material is the most im-

portant factor to account for the lower incidence of resistant strains in this study. The study has included a considerable proportion of patients without frank caseation or cavity, most of whom were treated for relatively indolent types of pulmonary tuberculosis. In such patients the incidence of resistant strains is shown to be extremely low (table 2).

The Influence of the Treatment Regimen Upon Sensitivity of M. Tuberculosis to Streptomycin

On the basis of experience in the joint Army, Navy, and Veterans Administration study (17) it was concluded that changes in the daily dose of streptomycin from 2.0 Gm. to 1.0 Gm. and, subsequently, even to 0.5 Gm. resulted in only minor differences in the incidence of streptomycin-resistant strains of tubercle bacilli. On all three regimens, resistant strains appeared after 30 to 45 days of treatment in an increasing number of patients until, after 120 days, about 70 per cent of positive cultures grew readily in concentrations of more than 10 γ per cc. The curves from this and other studies have suggested that the emergence of resistant strains was influenced far more by the duration of treatment than by daily dose of streptomycin, and that the percentage of resistant cultures was consistently low prior to approximately the sixth week of treatment. It was largely for this reason that a number of investigators have chosen to try shorter regimens, for example 42 or 45 days, in the hope that the resulting incidence of resistant strains would be materially reduced.

The number of cases in table 1 is undoubtedly too small to permit definitive conclusions from this study alone. It is apparent, however, that the change from the 120 day to the 42 day regimen has produced little change in the incidence of resistant strains encountered in this particular study. This is partly a consequence of the fact that increasingly resistant strains have appeared in some patients on the 42 day regimen after treatment was discontinued. If only those patients in table 1 known to have all cultures highly sensitive are considered, then Regimen C does have a distinct advantage. Even this advantage becomes slight, however, if patients whose sputum cultures became consistently negative during treatment (but whose last positive culture was highly sensitive) are included in the sensitive group. At the very least it must be concluded that reduction in the duration of streptomycin treatment from 120 days to 42 (or 45) days has been very much less successful than had been hoped in reducing the incidence of resistant strains of tubercle bacilli.

It is of incidental interest that there is limited evidence in the Laurel Heights study to suggest that, if tubercle bacilli are sensitive to streptomycin when sputum conversion occurs, they are likely to remain so. In addition to the patients whose bacilli must still be classed as indeterminate in tables 1 and 2, there were 8 other patients whose sputum and gastric contents became consistently negative for tubercle bacilli during treatment and whose cultures, prior to becoming negative, had all been sensitive to 0.5 or 1 γ. However, each of those 8 patients subsequently produced at least one or more positive specimens which permitted sensitivity again to be determined. These positive specimens occurred

from two to ten months after treatment had been completed. In every instance tubercle bacilli in the posttreatment specimen were sensitive to 10 γ per cc. or less, and in 6 of the 8 the bacilli were still sensitive to 0.5 or 1.0 γ. This experience is, however, different from that of Fisher (11) who found resistant strains in posttreatment specimens from approximately one-half of his patients who had obtained temporary bacteriologic conversion but had had recurrence of positive sputum three to twelve months later. (Fisher does not state whether or not resistant strains had already been observed in any of these patients prior to temporary sputum conversion.) At the present time one is compelled to regard sensitivity as indeterminate until the effect of the entire course of streptomycin treatment is definitely known.

The Influence of the Type of Pulmonary Tuberculosis upon Sensitivity of M. Tuberculosis to Streptomycin

Although the number of patients in table 2 is not large, the relationship between the type of tuberculosis being treated and the incidence of resistant tubercle bacilli appears far too close to be the result merely of chance. The patients in table 2 have been divided only into two broad categories of disease. Further subdivision would result in such small groups that the significance of any correlation observed might be questionable. It should be stated, however, that most (though not all) of the patients who had no frankly caseous or cavernous lesions were patients with the types of subacute or chronic pulmonary tuberculosis mentioned previously, namely those with disseminated nodular pulmonary tuberculosis and those termed "grumblers" (8). In these patients, the tuberculosis, while obviously active, was generally rather indolent in character. In contrast, the majority of the patients with frank caseation or cavity were treated for disease of much greater activity. In a considerable number, the tuberculosis was very active and rapidly progressive when treatment was started. It is believed that the activity of the tuberculosis was also a factor pertinent to the higher incidence of resistant strains in the caseous-cavernous group.

During the early experience with streptomycin in Great Britain, which was recently reported (21), treatment of pulmonary tuberculosis was confined to acute progressive tuberculosis of "bronchopneumonic" type, with or without cavity, in patients between the ages of fifteen and thirty. On the basis of sensitivity data from 41 such patients, it was concluded that there is a possibility of a relationship between the severity of the patients' clinical condition (high fever, large or multiple cavities) just prior to treatment and the development of high degrees of streptomycin-resistance. Also, Crofton and Mitchison (22), who participated in this cooperative British study, have analyzed data from 13 of their own patients. They found that resistant strains occurred earlier and that resistance reached higher levels in patients who did poorly during the early stages of streptomycin treatment than in patients who responded promptly and favorably.

In the Laurel Heights study, patients severely ill with the acute pneumonic forms of pulmonary tuberculosis have been relatively few. Included in table 2

are, however, 13 patients who were very ill with extensive progressive tuberculosis and high fever when streptomycin treatment was started. All these patients had extensive cavitation with highly active collateral disease, dense consolidations of at least lobar extent, or both. Among these 13 patients, resistant strains of tubercle bacilli emerged in 12. It is of further interest that in this same group there were 7 patients whose clinical response to streptomycin was negligible even during the first weeks of treatment. This experience was rarely encountered in other patients who were obviously ill clinically, but less severely so. The one severely ill patient who failed to discharge resistant strains of bacilli showed immediate and excellent clinical response to streptomycin treatment and eventually obtained almost complete resolution of an extensive tuberculous pneumonia. His sputum became consistently negative for tubercle bacilli after the third month of treatment and the sensitivity readings on all previous specimens had been 1.0 γ or less. In retrospect it is believed that caseation in this case was actually far less extensive than it was originally judged to be. In many patients, highly active tuberculosis, severe symptoms, extensive caseation or cavity, and poor response to treatment are such closely interrelated phenomena that it becomes difficult to analyze them separately. The experiences mentioned above suggest strongly that treatment of caseous or cavernous pulmonary tuberculosis with streptomycin for periods of 42 days or longer is, indeed, especially likely to result in the emergence of resistant strains of tubercle bacilli when the patient is severely ill with extensive and highly active disease. When, in addition, such a patient shows poor initial response to treatment, the eventual occurrence of resistant strains would seem to be almost certain.

Highly resistant strains eventually occurred, however, in several afebrile patients with chronic cavity and with only slowly progressive or chronically active collateral disease. Resistant strains also occurred in a considerable number of patients whose response to treatment, initially, was very prompt and very favorable. On the other hand, strains of bacilli uninhibited by 10 γ of streptomycin per cc. or less have occurred in none of several febrile patients treated promptly for acute exudative spreads of tuberculosis, but in whom frank caseation or cavity could not be detected. It appears from this study, therefore, that the presence or absence of frank caseation or cavity bears a far more consistent relationship to the emergence of resistant strains of tubercle bacilli than does the acuteness of the tuberculosis, the severity of symptoms, or the initial clinical response to treatment, when these factors are considered separately.

It cannot be inferred, however, that patients without frank caseation or demonstrable cavity in pretreatment roentgenograms are necessarily immune to the hazard of the emergence of drug-resistant strains of bacilli (table 2). The number of patients in this category is small, and, although highly resistant strains have not yet emerged in this particular group, strains less sensitive than those present prior to treatment have appeared. There is little reason to doubt that highly resistant strains also will eventually be encountered among patients of this type, though it is expected that the percentage will be small. Furthermore, it is obviously impossible in many instances to exclude completely the

presence of caseation or cavity on the basis of roentgenograms. In fact, even when *frank* caseation or cavity could not be demonstrated in the patients reported here, it is extremely likely that small foci of caseation often existed. Also, since tubercle bacilli were present in the sputum or gastric contents of all patients, it must be assumed that some degree of ulceration was present. In other words, it is well recognized that interpretations based largely on roentgenograms are bound to fall far short of strict pathological accuracy. Nevertheless, such interpretations have been made by phthisiologists long before the advent of streptomycin, and have long been used as an important guide to the management of patients. The practical value of such interpretations can scarcely be questioned.

The presence or absence of *frank* caseation or cavity, as judged from roentgenograms, has already been shown to influence greatly the therapeutic results obtained from streptomycin (7, 11, 17, 19, 23). It is not unreasonable that the incidence of resistant tubercle bacilli should also be influenced. It is in such lesions (and especially in the highly active and progressive ones) that tubercle bacilli are likely to be most numerous, that multiplication is likely to be most rapid, and that suppression by streptomycin is likely to be most limited.

In the present study the classification of patients on this basis actually presented little difficulty, although, naturally, borderline cases arose. The vast majority of patients judged to have caseous disease also had definite and clearly demonstrable cavity. In fact only 2 patients were placed in this category on the basis of caseation alone. In instances where small cavity was suspected but could not be definitely outlined, cavity was judged not to be present for purposes of classification.

Follow-up of patients treated at the Laurel Heights Sanatorium (7) has strengthened the conclusion that, when treatment by bed rest has failed, streptomycin alone is unlikely to produce sustained arrest of frankly caseous or cavernous pulmonary tuberculosis. In such cases, optimum results can be achieved only by the integration of streptomycin with appropriate forms of collapse therapy, resection, or other surgery. The observation that strains of tubercle bacilli resistant to streptomycin are especially likely to emerge, and that they often emerge rapidly in patients with frank caseation or cavity, further emphasizes the importance of an overall plan of therapy in such cases. It is only in this way that appropriate collapse therapy or other surgery can be applied promptly, early in the course of streptomycin therapy, before resistant strains emerge and prevent further benefit from the drug.

SUMMARY

1. Data on the sensitivity of tubercle bacilli to streptomycin have been presented in a series of patients treated with streptomycin for pulmonary tuberculosis. All were treated for 42 days or longer. Among these patients, the risk of streptomycin resistance proved to be related far more closely to the type of tuberculosis being treated than to the particular treatment regimen chosen. The presence or absence of frank caseation or cavity appeared to be the factor which most consistently influenced the liability to resistant strains.

2. In a group of 28 patients treated on a regimen of 1.8 or 2.0 Gm. of streptomycin daily for 120 days, resistant strains of tubercle bacilli eventually occurred in 8 (29 per cent). In a group of 47 patients treated on a regimen of 1.0 Gm. of streptomycin daily for 42 days (later revised to 20 mg. per kg. of body weight daily for 45 days), resistant strains eventually occurred in 13 (28 per cent). The number of patients in these two groups is too small to demonstrate in a definitive manner the relative effect of these regimens upon resistance. It is concluded, however, that a reduction in the duration of streptomycin treatment from 120 days to 42 (or 45) days has been considerably less successful than had been hoped in reducing the incidence of resistant tubercle bacilli.

3. A very impressive correlation has been observed between the incidence of resistant tubercle bacilli and the type of tuberculosis being treated. Resistant strains have emerged in a high proportion of patients who had frank caseation or cavity in pretreatment roentgenograms. In a group of 56 such patients, treated on the several regimens employed in this study, strains uninhibited by 10 γ of streptomycin per cc. of culture medium eventually occurred in 33 (59 per cent). In contrast (and regardless of the treatment regimen employed), strains uninhibited by 10 γ per cc. or less have occurred thus far in none of 31 patients who had no frankly caseous or cavernous lesions prior to treatment.

SUMARIO

Sensibilidad de los Bacilos Tuberculosos a la Estreptomicina. El Influjo de Varios Factores sobre la Emergencia de Cepas Resistentes.

1. Los datos presentados sobre la sensibilidad de los bacilos tuberculosos a la estreptomicina básanse en una serie de enfermos tratados con dicha droga por tuberculosis pulmonar. Todos ellos fueron tratados por espacio de 42 días o más. Entre ellos, el riesgo de la aparición de estreptomicinorresistencia resultó hallarse mucho más relacionado con la forma de tuberculosis en tratamiento que con el régimen terapéutico escogido. La presencia o ausencia de caseación o cavernas bien definidas pareció ser el factor que afectó más poderosamente la propensión al desarrollo de cepas resistentes.

2. En un grupo de 28 enfermos tratados con un régimen de 1.3 a 2.0 Gm. diarios de estreptomicina durante 120 días, se presentaron con el tiempo cepas resistentes de bacilos tuberculosos en 8 (29 por ciento). En un grupo de 47 tratados con un régimen de 1.0 Gm. diario de estreptomicina durante 42 días (modificado luego a 20 mg. diarios por kg. de peso durante 45 días), aparecieron con el tiempo cepas resistentes en 13 (28 por ciento). El número de enfermos en esos dos grupos es demasiado pequeño para demostrar definitivamente el relativo efecto de esos dos régimenes sobre la resistencia. Dedúcese, sin embargo, que la disminución de la duración de la estreptomicinoterapia de 120 a 42 (o 45) días ha surtido mucho menos efecto que lo esperado en lo tocante a reducir la incidencia de bacilos tuberculosos resistentes.

3. Observóse una correlación muy importante entre la incidencia de cepas tuberculosas resistentes y la forma de tuberculosis en tratamiento, apareciendo aquellas en una elevada proporción de los enfermos que mostraron manifiesta

caseación o caverna en las radiografías anteriores al tratamiento. En un grupo de 56 de estos enfermos, tratados con los varios régimenes empleados en el estudio actual, surgieron con el tiempo cepas que no inhibían 10 γ de estreptomicina por cc. de medio de cultivo en 33 (59 por ciento). En contraposición (e independiente del régimen terapéutico empleado), hasta la fecha no se han observado cepas que no inhibieran 10 γ por cc. o menos en ninguno de los 31 enfermos que no tenían lesiones francamente caseosas o cavernosas antes del tratamiento.

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COEXISTING DISSEMINATED COCCIDIOIDOMYCOSIS AND TUBERCULOSIS[1,2]

Report of a Case

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INTRODUCTION

Coccidioidomycosis has become a well recognized and clearly defined disease entity and many excellent studies have been made of its epidemiology, pathogenesis, clinical manifestations, and pathology (3, 8, 11). Although the parallelism of coccidioidomycosis and tuberculosis has been well studied (9), there have been only five reports (1, 4, 5, 6, 10) of the coexistence of the two diseases in the same individual. The present report is concerned with a sixth case, which is apparently the first observation of the association of *disseminated* coccidioidomycosis with tuberculosis and in which the dual etiology was proved both by culture and by animal inoculation. It is believed that this case manifests many epidemiologic, therapeutic, and pathologic features of unusual interest.

CASE REPORT

Clinical Course

The patient was a 34 year old white single enlisted man who had been well, except for a chronic "cigarette cough" until early August, 1942, when he was stationed at an Army Camp at Compton, California, some twenty miles south of Los Angeles. He had been stationed at Compton less than one month prior to the onset of symptoms, but had resided in San Diego, California, from November, 1941 to June, 1942. Initial symptoms consisted of a stiffness and soreness in the right anterior cervical region. In mid-August, because of this pain, a right lower tooth was pulled. Toward the end of August he noticed a gradual increase in size of the anterior cervical nodes, especially the right. This was associated with an aching in his neck which was aggravated by dorsiflexion and which kept him awake at night. During the month of August his cough increased in severity, and he developed general malaise, anorexia, and dysphagia.

The patient's past health had been excellent. He had measles and mumps in childhood, and lobar pneumonia at age 16. There is no history of tuberculosis or other chronic disease in himself or his family.

On admission to the 73rd Evacuation Hospital, on September 3, 1942, the patient appeared to be a chronically ill malnourished individual. He had a small superficial nodule with intact dermis and surrounding erythema on the forehead and another over the left scapula. There was a 3 cm. tender hard mass anterior to the right sternocleidomastoid muscle, as well as somewhat tender nodes varying in size from "shotty" to "pigeon egg" in both axillae and right and left anterior and posterior cervical chains. Severe pyorrhea was present. The remainder of the physical examination was negative.

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[1] From the Tuberculosis and Pathology Services of the Minneapolis Veterans Administration Hospital and the Departments of Medicine and Pathology of the University of Minnesota Medical School.

[2] Published with the permission of the Chief Medical Director, Department of Medicine and Surgery, Veterans Administration, who assumes no responsibility for the opinions expressed or the conclusions drawn by the author.

On admission the hemogram showed 3.7 million red blood cells per cu. mm., 80 per cent hemoglobin, 16,150 white blood cells per cu. mm., 60 per cent neutrophils, 10 per cent lymphocytes, 1 per cent basophils, 22 per cent eosinophils, and 7 per cent monocytes. The urinalysis was normal except for one plus albumin. The sputum was negative for acid-fast bacilli.

A chest roentgenogram showed metallic fragments present in the shoulder girdle area, at the site of an old bullet wound. There was also a soft 1 cm. infiltration in the right first interspace and two smaller adjacent infiltrations, "characteristic of tuberculosis." Examination of material obtained by biopsy of a lymph node and of one of the skin nodules revealed a "mycotic granuloma due to *Coccidioides immitis*."

Throughout the period spent at the 73rd Evacuation Hospital, the patient displayed an irregular fever with frequent elevations to 100° or 101° F. In September, 1942 a new skin nodule developed on the occiput and the old nodules were seen to be ulcerating and fungating.

The patient was transferred to Hoff General Hospital on September 27, 1942. His weakness and weight loss had progressed and he suffered from occasional spells of shortness of breath. His cough had become productive and he occasionally produced blood-streaked sputum. He first noticed a mild constipation at this time, which continued throughout his illness. There were no genito-urinary symptoms or signs.

Physical examination at this time revealed dirty ulcers, about 1.5 cm. in diameter, with raised red margins on the forehead, occiput and scapula. Enlarged nodes were found in the posterior cervical triangles, the left anterior cervical chain, and in the left axilla. The blood pressure was 110/74 mm. of mercury.

The hemogram remained essentially the same, with moderate leukocytosis and eosinophilia. The sedimentation rate varied from 72 to 117 mm. per hour. Urinalyses revealed a trace to one plus albuminuria as the only abnormality. Roentgenograms disclosed an increase in the infiltration in the right apex as well as new soft infiltrations in the left apex and left second interspace. Increased hilar shadows were also present. Subsequent roentgenograms showed progressive clearing so that by December, 1942, no active lesions were present. Roentgenograms of the skull and shoulder regions were negative.

Examination of material obtained by biopsy of the skin ulcer again showed a granulomatous lesion containing spherules of *Coccidioides immitis*. Three sputum examinations for acid-fast organisms were negative, but *Coccidioides immitis* was reported present in the sputum. Skin tests with coccidioidin in dilutions of 1:1,000, 1:100, and 1:10 were all negative. Complement fixation tests gave a one plus reaction at 1:64 dilution, and a positive precipitin test in a dilution of 1:40.[3] Complement fixation tests for *Blastomyces*, performed by the Department of Medicine, Duke University School of Medicine, were negative. Material from the forehead lesion sent to Dr. C. E. Smith of Stanford University School of Medicine, cultured on Sabouraud's agar, blood agar, and a special differential *Coccidioides* medium, grew a white cottony fungus which was injected into two mice. Both mice died and typical double-contoured endosporulating spherules were found in the lungs. These were cultured a second time and again produced a cottony white fungus. No *Blastomyces* were found.

The patient's course in Hoff General Hospital was at first irregularly febrile, with frequent daily elevations of temperature to 100 to 101° F. Therapeutic trials of sulfathiazole were made on two occasions, but were discontinued each time because of fever and dermatitis. Fever therapy with typhoid vaccine was tried without attaining a significant temperature response. Malaria fever therapy was then instituted, during which the patient developed seven chills with no change in the clinical course. However, the total length of time in which the temperature was over 102° F. was less than twenty hours. Also during

[3] All serological tests, unless otherwise indicated, have been performed by Dr. C. E. Smith, of the Stanford University Medical School, Department of Public Health, and the Commission on Acute Respiratory Diseases of the Army Epidemiological Board.

this period the local skin lesions were excised and treated with roentgen therapy. It was noted that gradually the old skin lesions healed completely although a new papule developed on the ramus of the jaw. The lesions demonstrable on the chest roentgenograms cleared and the temperature gradually became normal.

On January 16, 1943, the patient was transferred to McCloskey General Hospital where, upon admission, it was noted that he felt well, had no cough and no fever. Examination disclosed the well healed scars of the old lesions on the forehead, scapula, occiput, and sternum, with an active crusted lesion present on the ramus of the jaw. Otherwise the physical examination revealed no relevant abnormalities.

Roentgenograms taken at the time of admission showed pleural thickening in the right apex and an infiltration behind the right clavicle, which did not change on subsequent examinations through July 10, 1944. The apex of the left lung was clear. Films of the cervical spine were negative except for the presence of hypertrophic changes about the vertebral bodies. There was no essential change in the red cell count or hemoglobin. The white blood cell count ranged between 5,950 and 9,000 per cu.mm. with 7 to 21 per cent eosinophils. A skin test for tuberculosis using Purified Protein Derivative (first strength) was positive in May, 1943, but thirteen separate sputum examinations for acid-fast bacilli were negative. Except for one positive culture in March, sputum cultures for *Coccidioides immitis* were all negative during 1943. One feces culture for *Coccidioides* was reported positive in February, 1943. A guinea pig inoculation of material from a skin lesion was positive for *Coccidioides immitis* in June, 1943. Biopsies of the skin lesions were reported to show coccidioidal granuloma in February and May, 1943.

Skin tests with coccidioidin 1:1,000 and 1:100 were negative in March and October 1943. In December 1943, complement fixation tests showed a two plus reaction in the dilution of 1:64. The precipitin test was negative.

During 1943 the patient had only occasional episodes of low grade fever. In February, 1943, new lesions appeared on the sternum and perineum, and these, as well as active lesions on the nose, forehead, and left lower mandible were treated with surgical cautery, with temporary benefit.

From May, 1943 to January, 1944, the patient received several courses of saturated solution of potassium iodide in doses which ranged between 3.0 and 8.0 cc. administered three times daily. Although some healing of the lesions occurred during this therapy, they invariably relapsed when the iodide was discontinued. Moreover, the iodide therapy was accompanied by a transient enlargement of the thyroid.

On September 11, 1944, the thyroid was noted to be normal to palpation, the skin lesions were quiescent, and the patient was given a medical discharge from the Army.

The patient returned to his home in Minnesota, but was unable to return to work. A note in his record, dated September 18, 1944, states that there was no active evidence of disease.

The patient was admitted to the Minneapolis Veterans Administration Hospital in January, 1945, at which time it was noted that he felt fairly well but was still very weak. He had been taking potassium iodide irregularly and had noted that occasionally his thyroid would "swell up." Except for his appearance of malnutrition, physical examination revealed no evidence of active disease.

Roentgenograms at this time demonstrated old well-defined infiltrates in the right apex, right fourth anterior interspace,and left base, as well as a diffuse soft lesion in the left anterior second interspace (figure 2A). He was discharged after this examination.

A letter dated February 25, 1945, from his physician states that the patient had been unable to work because of marked fatigue. An active granulomatous ulcerating lesion had appeared on his nose for which he was taking potassium iodide, 5.0 cc. daily, without effect.

Roentgenograms in May, 1945 showed some increase in lesions in the right and left subapical regions. The urinalysis was negative and the hemogram was normal. One sputum examination was negative for *Coccidioides immitis*. The basal metabolism rate was plus 27. Serologic tests for syphilis were negative.

By July, 1946 his nasal lesion was noted to have increased in size, but the chest roentgenographic findings showed no change. There had been no other change during the previous year.

The patient was admitted to the Minneapolis Veterans Administration Hospital for the last time on December 11, 1947. During the preceding six months he had lost an additional 15 pounds of weight, had become weak to the point of prostration, and had developed a marked anorexia, as well as a cough productive of small amounts of sputum. In September, 1947 he had first noticed a sore throat which now made it difficult for him to swallow. He was taking potassium iodide, 3.5 cc. daily and stated that he had tried a few times during the preceding year to discontinue this medication, but that each time he did so the nasal lesions became worse.

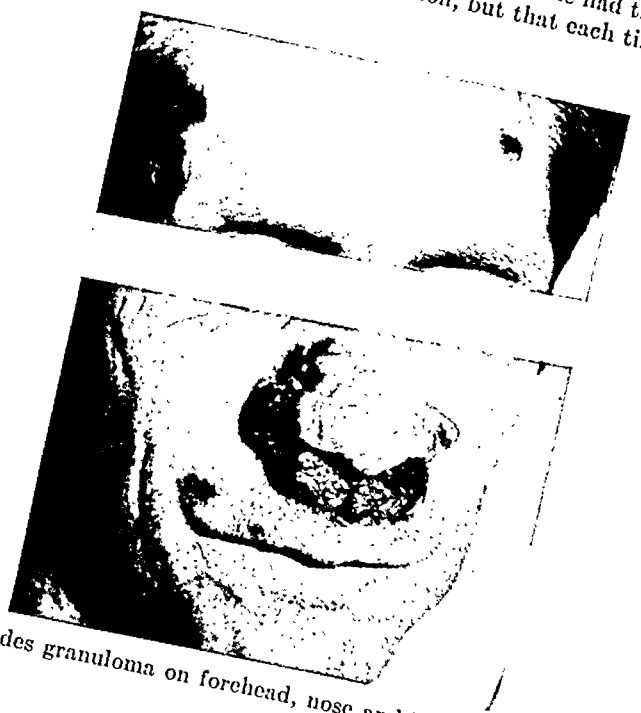

FIG. 1. A coccidioides granuloma on forehead, nose and lip (photographed December, 1947).

On physical examination the temperature was 99.4° F., the pulse 104 per minute, and the respirations 18 per minute. The blood pressure was 90/60 mm. of mercury. It was noted that he was a poorly nourished, chronically and acutely ill, cachectic white male. There were granulomatous ulcerating skin lesions, smaller than a dime, on each side of the forehead. A large fungating, crusting, and oozing granulomatous lesion covered both nares, both alae nasae, the tip of the nose, and extended onto the upper lip (figure 1). Well healed scars were noted at the sites of the old cauterized lesions. The pharynx and tonsils were covered by ulcerating and granular lesions that completely replaced the normal mucosa. One almond-sized, freely movable nontender node was found in the right posterior cervical chain, and many smaller nodes were palpable in the neck, axillae, and groin. The left lobe of the prostate was enlarged, firm, and tender, and the left seminal vesicle was enlarged and tender. The remainder of the physical examination was negative.

The hemogram on admission was normal except for an eosinophilia of 17 per cent. The white blood cell count rose gradually to 18,300 cells per cu.mm. with a predominant polymorpho nuclear leukocytosis.

Urinalysis on admission revealed a four plus albumin reaction. The microscopic field was "solid" with white blood cells, and occasional red blood cells.

Roentgenograms of the long bones and skull were all normal in December, 1947. An intravenous pyelogram in December was also normal. Roentgenograms of the chest on admission (figure 2B) showed, in addition to the previously described metallic densities, diffuse exudative and nodular infiltrations radiating outward from the hilum through both lung fields, characteristic of a recent bronchogenic dissemination of tuberculosis or of a widespread mycotic infection. In January, 1948, roentgenograms showed clearly, for the first time, evidence of bilateral cavitation chiefly involving the upper lobes. On February 7, 1948, in addition (figure 2C), a diffuse miliary dissemination was seen throughout both lung fields.

Results of serologic and bacteriologic examinations pertaining to coccidioidomycosis and tuberculosis are given in table 1.

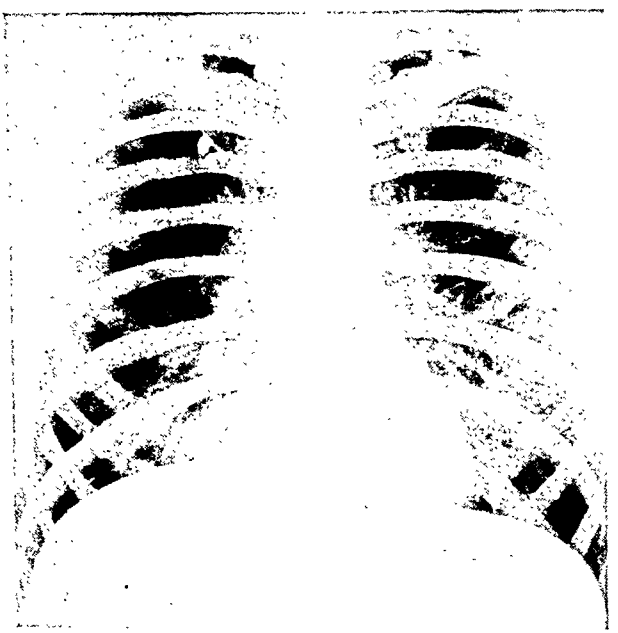

FIG. 2A. Roentgenogram in 1945, three years after onset, showing infiltration in left third interspace, right apex and right base.

Coccidioidin skin test (1:1,000 and 1:100), each performed twice, were negative. Tuberculin skin test (PPD #1 and PPD #2) were negative. Heterophile antibody and cold agglutination tests were negative.

Examination of material obtained by biopsies of the skin lesion and of the tonsil showed definite granulomatous changes with true tubercle formation. In the material obtained from the skin many typical double contoured spherules containing endospores were seen. Special staining failed to show acid-fast organisms in the skin lesion. Fewer spherules were seen in the tonsilar lesions and here many acid-fast organisms were demonstrated. Sectioned and smeared material from the sternal marrow and from the sputum showed no organisms or granulomas.

Seven relatives of the patient, with whom he had been in close association since discharge from the Army, including the patient's mother, were skin tested with coccidioidin in dilution 1:100. All were negative at twenty-four and forty-eight hours.

Cerebrospinal fluid examination on December 16, 1947, was negative. On January 15, the c.s.f. sugar was 53 mg. per 100 cc., the protein was 62 mg. per 100 cc. and microscopic examination disclosed 88 white blood cells per cu. mm., of which 90 per cent were mono-

nuclears. A first zone type of colloidal gold curve was present. Essentially similar cerebrospinal fluid findings were present on February 12.

On admission the patient was febrile with daily temperature elevations to 99.6° to 100° F. The fever subsided after one week but recurred intermittently. At the time of admission, the patient was placed on penicillin, 40,000 units every three hours, with no benefit.

Because of the marked discomfort in the pharynx, cauterization with 25 per cent trichloracetic acid was tried on two occasions without relief. By December 30, pain from the ulcerated pharynx had become so severe that the patient was able to swallow liquids only with difficulty and solids not at all. At this time he was given streptomycin 0.5 Gm. twice daily intramuscularly, which was continued until his death. Within two days the

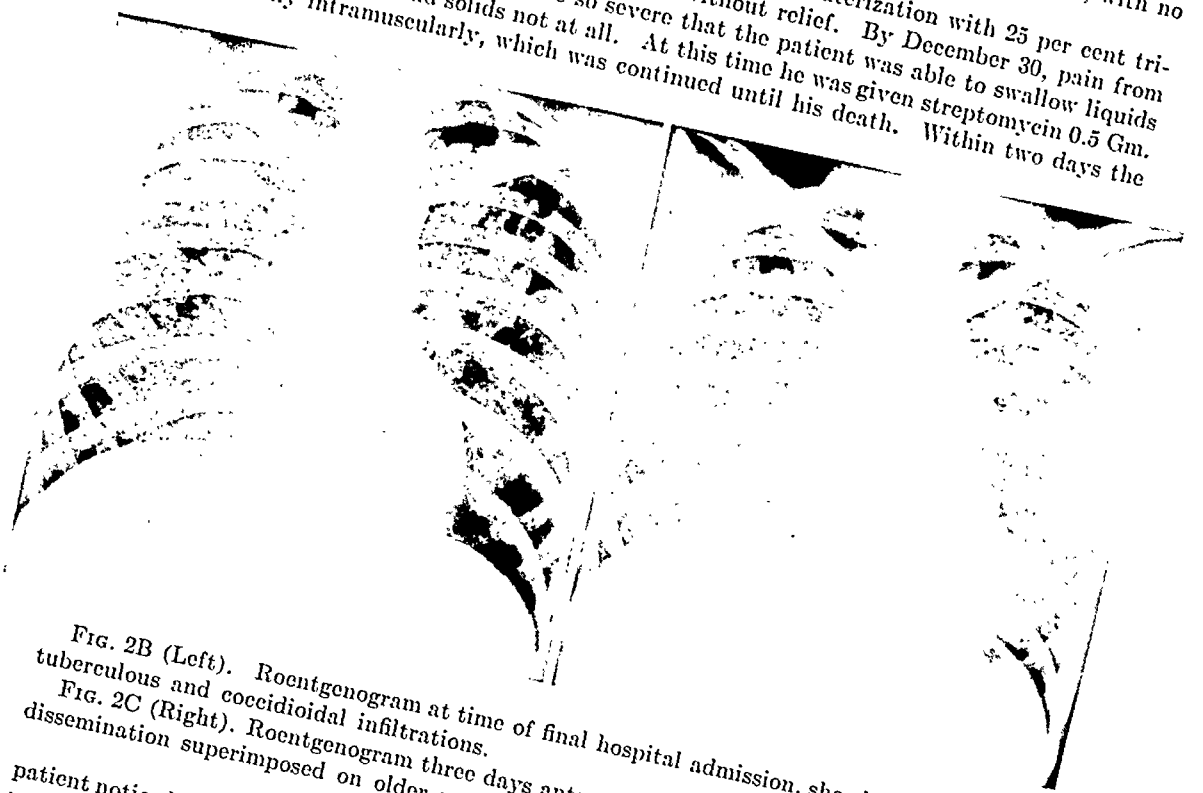

FIG. 2B (Left). Roentgenogram at time of final hospital admission, showing extensive tuberculous and coccidioidal infiltrations.

FIG. 2C (Right). Roentgenogram three days antemortem, showing miliary coccidioidal dissemination superimposed on older processes.

patient noticed subjective throat improvement. The pharyngeal and tonsilar ulcers healed until, by February 2, the pharynx was covered by an intact mucosa.

Despite the definite healing of the pharynx, the patient's general condition gradually became worse. He complained of headache and some stiffness of the neck for the first time on January 6, and the skin lesions were noted to be larger, more swollen, and oozing considerably. On January 12, new active skin lesions appeared on the lower face and chin. The following day calciferol, 1.0 cc. (50,000 units of Vitamin D_2) orally in three doses per day was started. After two days this was changed to drisdol in propylene glycol (50,000 units of Vitamin D_2) 0.5 cc. intramuscularly twice daily. This was continued until his death. On January 14, for the first time there was definite objective neck rigidity and a positive straight leg raising test. During the ensuing three weeks, symptoms and signs of meningitis continued to wax and wane, but were never very marked.

From January 20 through January 23, the patient received 4.0 cc. of saturated solution of potassium iodide daily. On January 26, the skin granulomas were noted to be less elevated and were beginning to crust over, but within four days they had become more active, only to subside again. By February 7, the edema, erythema, and exudate had de-

creased markedly. No new skin lesions appeared after January 12, and the dermatitis never again became as active as it had been in mid-January.

On February 7, the patient rapidly became more dyspneic and weaker. The respirations were shallow, and the rate was 30 per minute. The pulse rose to 148 and the rectal temperature to 101° F. The pulse, temperature, and respiratory rates remainded elevated. Fine scattered rales and occasional wheezes were heard throughout the chest. The blood pressure remained at 84/60 mm. of mercury. A chest roentgenogram revealed the presence of a miliary dissemination. The patient became cyanotic and oxygen was given. On February 9, a pleural-pericardial friction rub was heard, and three days later a pleural

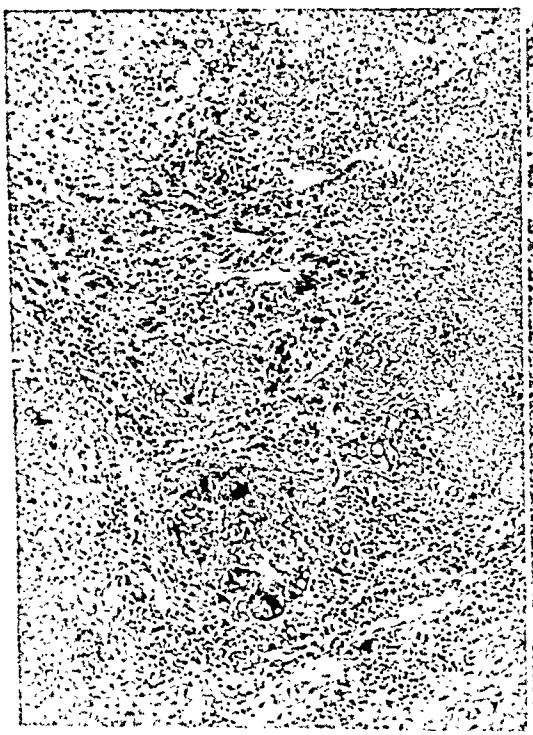

FIG. 3 (Left). A typical coccidioides granuloma from the adrenal cortex containing epithelioid cells, giant cells and coccidioides spherules.

FIG. 4 (Right). A tubercle of the lung with occasional coccidioides spherules at the periphery.

friction rub was noted. The patient became rapidly weaker and expired on February 13, 1948.

Autopsy Findings

Autopsy was performed six hours after death. The body was that of a well developed, poorly nourished white adult male weighing 110 pounds and measuring 170 cm. in length. At the external nares and covering most of the upper lip below the nose, there was a large, irregular, slightly raised granulomatous lesion extending a few millimeters into the external nares, onto both alae nasae, and the tip of the nose. It was covered by a thick, yellowish brown, firm, nodular crust and had a well defined though irregular margin, without tumescence. The surrounding skin showed no abnormalities. There were similar smaller lesions at the following sites: two on the right side of the upper lip, both one-half cm. in diameter; one on each side of the forehead, symmetrical, and measuring each one cm. in diameter; and one on the tip of the left index finger at the border of the nail.

The peritoneal surfaces were smooth and glistening. There were approximately 50 cc. of clear yellow fluid in the peritoneal cavity. The pleural cavities contained no fluid. There were numerous firm fibrous adhesions between the visceral and parietal pleural surfaces over the middle and upper lobes of the right lung and a few fibrous adhesions at the apex of the left lung. The pericardial cavity contained 30 cc. of clear yellow fluid. The pericardial surfaces were smooth and glistening. The heart showed no gross abnormalities. The lungs together weighed 2,125 Gm. There was a moderate thickening of the pleura over the upper and middle lobes of the right lung. A few pin point nodules were seen to be scatterd irregularly over the pleural surfaces of both lungs. The bronchi contained thick, pinkish fluid but in other respects showed no abnormalities. The parenchyma of the lungs was quite firm on palpation, and crepitation was reduced throughout all the lobes. On section, the entire parenchyma of both lungs had a grey solid appearance, imparted to it by closely approximated yellowish grey, punctate nodules ranging in diameter from one-half to one mm. Many of these nodules were confluent. There were several yellowish caseous-appearing nodules measuring from 3 to 5 mm. in diameter, chiefly in the apical portion of the right lung, but also in the mid-posterior portion of the left upper lobe and the upper portion of the left lower lobe. Three small cavities were seen, located respectively in each of the areas containing caseous nodules. The largest of these, measuring 2 cm. in diameter, was in the mid-posterior portion of the left upper lobe. The other two measured one cm. and one-half cm. in diameter. The cavities had fibrous walls measuring one to two mm. in thickness and contained very viscid yellow thick fluid.

The spleen weighed 225 Gm. Its parenchyma was dark purple and diffusely studded with punctate white nodules. The liver weighed 2,900 Gm. and showed no gross abnormalities. The right adrenal gland weighed 20 Gm. It was firm and slightly rubbery on palpation. The entire central and anterior portion of the gland was replaced by lobulated masses of yellowish grey, cheesy looking material. Only a small portion of the cortex remained intact. The left adrenal gland was of normal size, but had similar replacement of its central portion. In the lower portion of the ileum were numerous ulcerations which measured one-half by one cm. in diameter, with axial polarity. The margins were ragged and the bases yellowish and necrotic; the serosal coverings of these ulcerated areas were reddened and nodular. The kidneys weighed a total of 420 Gm. The parenchyma of both kidneys was diffusely studded with small white punctate nodules. In the left kidney, in the medullary portion, near one of the papillae, there was a small cystic cavity measuring one cm. in diameter. It had a smooth, thin, fibrous wall and was filled with thick greenish-yellow fluid. It did not appear to communicate with the pelvis of the kidney. The prostate was enlarged and boggy. All the ducts were dilated, giving the gland a cystic appearance, and were filled with thick yellow, creamy fluid. The seminal vesicles were moderately enlarged and bulging. They were filled with thick cheesy, greyish yellow material. The testes and epididymides showed no abnormalities.

The mediastinal, hilar, anterior cervical, and axillary lymph nodes were enlarged measuring one to two cm. in diameter. They had a rubbery firm consistency and pink homogeneous parenchyma.

On examining the brain, the leptomeninges in both cerebellopontine angles were found to be thickened, milky, and adhesive. A few small punctate, milky exudates were found along the cerebral vessels, especially in the Sylvian fissure. There were scattered punctate, granular nodules on the ventricular surfaces.

The parenchyma of the lung, on microscopic examination, was seen to be extensively disrupted by focal and diffuse granulomatous involvement, which generally had a lobular distribution and frequently gave the appearance of a granulomatous pneumonia. The interalveolar septae were largely preserved. The alveoli were filled with epithelioid cells and multinucleated giant cells, with shreds of necrotic debris, small lymphocytes, and plasma cells. Around the periphery of these areas, the alveoli were filled with macrophages, a few small lymphocytes, and contained an occasional giant cell. Numerous spherules of *Coccidioides immitis* were seen in these areas; most of these were contained in multinucleated giant cells; a few were found in macrophages.

Scattered throughout the sections of the lungs examined, in addition to the above type of granuloma, there were also seen numerous small and large masses of compact caseous necrotic material surrounded by epithelioid cells, multinucleated giant cells of the Langhans' type, and small lymphocytes. The pulmonary parenchyma was completely destroyed in these areas. Acid-fast stains revealed that many of these latter contained acid-fast bacilli in the zones of epithelioid and giant cell reaction. An occasional *Coccidioides immitis* cell was seen at the periphery of one of the small lesions of this type.

Sections of one of the cavities in the lungs revealed it to be lined by a loose fibrous capsule and to be filled with a dense mononuclear cell exudate, chiefly composed of plasma cells, but also containing macrophages and multinucleated giant cells in a loose fibrous tissue network. There were numerous coccidioidal organisms seen, most of which were in the giant cells and macrophages. The central portion of the cavity was seen to contain basophilic necrotic debris in loose clumps.

The spleen contained numerous small scattered granulomas, which contained large multinucleated giant cells, a few small lymphocytes, and plasma cells, and shreds of necrotic debris at their centers, with surrounding epithelioid cells and concentric rings of loose fibrous tissue. Occasional granulomas also contained neutrophils and eosinophils. Spherules of *Coccidioides immitis* were found in the granulomas, usually within giant cells.

The liver lobules contained a few of the same type of small granulomas, with organisms of *Coccidioides immitis*. The portal spaces were diffusely infiltrated by lymphocytes, plasma cells and a few eosinophils, and contained an occasional multinucleated giant cell.

The entire central portion of the right adrenal gland was replaced by large masses of caseous necrotic material, with only a thin rim of intact cortical cells. This material was surrounded by a broad zone of cellular reaction composed of masses of polymorphonuclear leukocytes, some small lymphocytes and plasma cells, and scattered eosinophils. The neutrophils lay in dense pockets resembling small abscesses. There were a great number of coccidioidal organisms in both the caseous area and the surrounding reaction zone. Some of these were filled with or were discharging endospores which lay in proximity to small abscesses. Most of the organisms in the cellular zone were within giant cells. The left adrenal gland was similarly involved.

The ulcers in the ileum extended to or through the muscularis mucosa, had necrotic bases and undermined edges. Under the necrotic base was found a broad zone densely infiltrated by small lymphocytes and plasma cells and containing scattered tubercles with caseous necrotic centers. No coccidioidal organisms were seen but special staining revealed the presence of numerous acid-fast bacilli.

The small cavity in the left kidney contained clumps of slightly basophilic necrotic debris, many neutrophils, and a few mononuclear leukocytes. Spherules of *Coccidioides* lay chiefly in giant multinucleated or large mononucleated cells in the peripheral portions of the cavity. The wall of the cavity was composed of a dense zone of polymorphonuclear leukocytes. Scattered throughout the remainder of the cortex and medulla of both kidneys were numerous small granulomas resembling those in the spleen, but containing a greater number of eosinophils and spherules.

The ducts and acini of the prostate contained numerous abscesses filled with neutrophils and spherules. Many of the spherules contained endospores. The abscesses were surrounded by a zone of chronic granulation tissue, containing chiefly plasma cells, some lymphocytes, eosinophils and occasional spherules.

The seminal vesicles were distended with caseous necrotic material and their epithelial lining was largely destroyed. The necrotic masses were surrounded by zones of epithelioid cells and lymphocytes extending into the surrounding stroma. No organisms of *Coccidioides* were seen, but numerous acid-fast bacilli were demonstrable with appropriate staining. The distal portions of the ducti deferens were similarly involved, but the epididymis was free of involvement.

The thyroid gland contained a few small coccidioidal granulomas located within the interlobular connective tissue, which was partially hyalinized. The lobules were free of cellular infiltrate.

Although some of the hilar nodes were uninvolved, many were filled with coccidioidal granulomas around which were dense bands of partially hyalinized connective tissue.

The lesion on the tip of the left index finger was a dense subepidermal inflammatory reaction composed of plasma cells, lymphocytes, eosinophils, and multinucleated giant cells, many of which contained *Coccidioides* spherules. There was one large subepidermal vesicle containing acidophilic fluid, many degenerating neutrophils, and a few spherules.

The leptomeninges were diffusely thickened and chronically inflamed, with numerous granulomas containing coccidoidal organisms. The same type of lesions were found in the

TABLE 1

Mycology and bacteriology of material obtained before and after death

	SPUTUM				PROSTATE			URINE				SPINAL FLUID			
	Tuberculosis smear (1)	Tuberculosis Culture	Tuberculosis Guinea pig	Coccioides Culture (2)	Tuberculosis smear	Tuberculosis Culture	Coccioides culture	Tuberculosis smear	Tuberculosis Culture	Tuberculosis Guinea pig	Coccioides culture	Tuberculosis smear	Tuberculosis Culture	Tuberculosis Guinea pig	Coccioides culture
12/15/47				+											
12/19/47	IV	+		0											
12/21/47	I														
12/22/47				0											
12/23/47		+	+	0				+			0				
12/26/47											+				
1/ 6/48								+	+						
1/ 7/48	VI	+		+				0	+		+ (4)				
1/12/48	II	+						0	+						
1/15/48								0	+	+					
1/19/48	I			+											
1/22/48											0	0		0	
1/28/48	0							0	0						
2/11/48	0	+		0	0					+	0	0	0		0
2/12/48						+								+ (5)	
2/13/48						+ (3)									

(1) All direct smears reported in Gaffky units.

(2) Unless otherwise indicated, all "positive" coccioides culture reports are based only upon gross examination of Sabouraud's medium culture.

(3) In addition to this "positive" culture obtained at autopsy, typical "positive" cultures were also reported from right lung, left lung, and kidney abscess. Only the last was confirmed by guinea pig inoculation.

(4) "Positive" culture, guinea pig negative.

(5) "Positive" culture, guinea pig positive.

choroid plexus and in the ependyma. Scattered granulomas containing a few organisms were also found in the white matter of the cerebral hemispheres and the pons. Acid-fast stains failed to disclose the presence of tubercle bacilli in the brain or meninges.

Acid-fast bacilli, resembling the tubercle bacillus were found only in the lungs, gastrointestinal tract, and seminal vesicles. Despite numerous attempts to demonstrate them, acid-fast organisms were not seen in any other organ.

Mycology and Bacteriology

The methods of identification of the *Coccidioides immitis* in this case were similar to those described by Dickson (2), and by Smith (8). Cultures obtained antemortem and postmortem, and their results, are shown in table 1. All of

these cultures were grown on Sabouraud's agar at room temperature. Positive cultures had an abundant white cottony growth in four to five days. From five to seven days later the growth had acquired a more fluffy appearance. Some of the growth was removed at this time from one representative culture (kidney abscess) and suspended in saline. On microscopic examination numerous chlamydospores were seen. This suspension was then injected intraperitoneally into a male guinea pig. After three weeks the guinea pig was given an intradermal injection of coccidioidin, 1:1,000. The test was read in thirty-six hours and was positive. The animal was then sacrificed. Typical spherules were recovered from an abscess of a testicle on direct smear. This process was repeated on cultures of the urine and cerebrospinal fluid, with positive results in the latter. Both the testicles and spleen of the test animal were found to contain granulomas filled with organisms of *Coccidioides immitis* on histologic examination.[4]

A record of the cultures for *M. tuberculosis* is also presented in table 1. Material for culture was concentrated and poured onto a slant of Petragagni's medium, then incubated at 37.5° C. for three to eight weeks. Positive cultures had yellow, flaky, rough colonies which were as a rule identified on macroscopic examination. Questionably positive cultures were further investigated by examination of a direct smear of the cultures. The guinea pigs were inoculated subcutaneously in the abdomen and intramuscularly in the groin with spinal fluid and sputum concentrates. After two months, the animals were sacrificed and examined for the presence of gross miliary tubercles in the spleen and liver. Using this method, only the sputum was found to contain *M. tuberculosis*.

A culture on Sabouraud's medium, obtained antemortem from the skin lesions, was sent to Dr. Robert M. Allen of the University of Nebraska College of Medicine. He injected the culture into mice and found coccidioidal spherules on histologic examination.

DISCUSSION

Histologic evidence of coccidioidal infection in this case was found in the lungs, liver, spleen, adrenals, kidneys, prostate, thyroid, meninges, brain, skin, hilar and mediastinal lymph nodes, and tonsils. There were two types of histologic response to the *Coccidioides* invasion. The first type, a granuloma composed of multinucleated giant cells surrounded by a loose net of epithelioid cells was seen throughout the organs involved. The second type, a typical suppurative lesion, was seen chiefly in the prostate and kidney. A gradation between these two types of tissue reaction was seen in the adrenals.

The granulomas in which *Coccidioides immitis* predominated differed from the tuberculous granulomas in their lack of caseation and less compact structure of the epithelioid cells (figure 3). The giant cells of the coccidioides granulomas did not show the regular peripheral crescentic placing of the nuclei as found in

[4] It may be noted that, at least in this laboratory, the testicle of the male guinea pig proved the most frequent site of growth of the coccidioidal organism after injection of culture suspensions. No growth was found after injection of culture suspensions into female guinea pigs.

the Langhans' giant cell of tuberculosis. Most of the cavities found in coccidioidal lesions were areas of liquefaction in a granuloma containing an unusual number of neutrophils. In contrast, the areas in which tubercle bacilli were found were typical caseous tubercles with Langhan's giant cells and epithelioid cells (figure 4).

This case was typical of disseminated coccidioidomycosis, in that although the prostate was involved and culture of the urine was positive, no involvement of the renal pelves or ureters was encountered. Similarly, no involvement of the bowel with *Coccidioides* was noted, although many typical tuberculous ulcers were seen.

Apparently, in this case the tuberculosis and coccidioidomycosis were coincidental. The tuberculosis was found chiefly in the lung and had disseminated only to the tonsil, bowel, and seminal vesicles. Except for the last, there was no involvement which suggested hematogenous dissemination. The coccidioidomycosis, however, had disseminated widely and was probably the cause of death.

Of special interest is the fact that, although the coccidioidin skin test was negative from the first observation in 1942, the tuberculin skin test was positive as late as May, 1943. This would indicate that, at the time the coccidioidomycosis became disseminated, at least some of the patient's immune responses were functioning normally and that his anergy to coccidioidin was an isolated phenomenon. Terminally, as both diseases became generalized, the patient also lost his ability to react to tuberculin.

Although epidemiologists have failed to demonstrate the contagiousness of human coccidioidomycosis, Rosenthal and Routien (7) have recently produced pulmonary coccidioidomycosis in guinea pigs by the intratracheal inoculation of sputum, pus, and other material from cases of human coccidioidomycosis. In view of these writers' conclusions that sputum and other material "can be infective through the respiratory route," it is interesting to note that 7 individuals who had lived in close association with our patient over a period of thirty-nine months demonstrated neither positive skin tests nor any other evidence of coccidioidomycosis.

The present case illustrates again the lack of any curative therapeutic attack upon disseminated coccidioidomycosis. During the course of the disease, sulfonamides, penicillin, streptomycin, fever therapy, local cautery, potassium iodide, and massive doses of vitamin D_2 were all tried. The first three were of no value. The fever therapy may have been of some temporary benefit, as was the local cautery. Although numerous other cases have been placed on potassium iodide with no benefit, this medication apparently produced a definite retarding effect on the progress of our patient's coccidioidomycosis. The effect of the iodide on the tuberculous processes is more obscure. Although potassium iodide was begun in May, 1943 and continued intermittently until December, 1947, acid-fast bacilli were not noted in the patient's sputum until the latter date. At this time iodide therapy was discontinued. Four cc. of potassium iodide daily were administered from January 20 through January 23, 1948, with no

obvious change in the course of the tuberculosis. It is not possible on the basis of this evidence to draw any definite conclusion as regards the influence of potassium iodide on tuberculosis.

Neither can conclusions be drawn in regard to the use of massive doses of vitamin D_2 in coccidioidomycosis of the skin. However, the coccidioidal lesions cleared sufficiently during the period of administration of calciferol to warrant further clinical trial of the drug.

Perhaps the most interesting observation was that of the differential effect of streptomycin, which it is believed definitely aided the healing of the patient's pharyngeal tuberculosis, while his coccidioidomycosis grew worse.

SUMMARY AND CONCLUSION

A case of disseminated coccidioidomycosis which lasted for six years and was complicated by active tuberculosis is presented. The two diseases seemed to occur coincidentally, the individual coccidioidal granulomas being devoid of acid-fast bacilli, and the tubercles containing only a few *Coccidioides* spherules at the periphery. The coccidioidomycosis terminated in a miliary dissemination throughout the body, including the brain and prostate, as well as the more usual sites, but did not involve the bowel or renal pelves. The tuberculosis presented a definite "secondary" or "adult" type of distribution in the lungs, pharynx, and bowel, as well as a hematogenous dissemination to the seminal vesicles.

Streptomycin was of definite value in controlling the tuberculosis but did not affect the coccidioidomycosis, whereas potassium iodide and vitamin D_2 in massive doses seemed to decrease some manifestations of the coccidioidomycosis. It is suggested that the latter drugs be given further trial in cutaneous coccidioidomycosis.

SUMARIO Y CONCLUSIONES

Coccidioidomicosis Difusa Coexistente con Tuberculosis

El caso presentado es de coccidioidomicosis difusa de seis años de duración, complicado por tuberculosis activa. Las dos enfermedades aparentemente coincidieron en su aparición, hallándose los distintos granulomas coccidioideos desprovistos de bacilos ácidorresistentes y conteniendo los tubérculos apenas algunas esférulas de *Coccidioides* en la periferia. La coccidioidomicosis terminó en difusión miliar por todo el cuerpo, incluso el cerebro y la próstata así como los sitios más habituales, pero sin afectar el intestino o las pelvis renales. La tuberculosis mostró una forma de distribución "secundaria" o "adulta" bien definida en los pulmones, faringe e intestino, así como difusión hematógena a las vesículas seminales.

La estreptomicina resultó de valor decidido en lo relativo a cohibir la tuberculosis, pero no afectó la coccidioidomicosis, en tanto que el yoduro de potasio y la vitamina D_2 a dosis masivas parecieron atenuar algunas manifestaciones de la última. La última droga podría muy bien ser objeto de nuevas pruebas en la coccidioidomicosis cutánea.

Acknowledgment

Dr. William B. Tucker, Chief of the Tuberculosis Service, Minneapolis Veterans Administration Hospital, and Dr. C. E. Smith of the Department of Public Health and Preventive Medicine, Stanford University Medical School, have given invaluable aid in the preparation of this paper. To the members of the staff of the 73rd Evacuation Hospital, the Hoff General Hospital, and the McCloskey General Hospital, we are indebted for the excellent study they made of the early course of this case.

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VIABILITY OF TUBERCLE BACILLI IN EMBALMED HUMAN LUNG TISSUE[1]

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INTRODUCTION

In the course of a study of tuberculin conversions occurring among medical students at the University of Rochester School of Medicine, Rochester, New York, it was found that the majority of conversions which occurred during the four years of the medical curriculum occurred during the first and second academic years. By far the majority occurred during the second year and it was concluded that these were due to contact with tuberculosis autopsy material (1). It was also noted, however, that a certain number of conversions took place during the first academic year when there was no known contact with cases of active pulmonary tuberculosis or with tuberculosis autopsy tissues. It seemed of interest, therefore, to investigate the possibility of infection of students from the tissues of cadavers used for dissection purposes in courses in anatomy. Dissection cadavers are always embalmed and it has been generally assumed that embalming suffices to kill any tubercle bacilli present. However, a search of the literature revealed no study covering this particular point. With this in mind it was decided to investigate the question of the viability of tubercle bacilli in embalmed cadavers which had known pulmonary tuberculosis. This paper reports the results of such a study.

METHOD

A letter outlining the problem and the reason for the investigation was sent to approximately eighty departments of anatomy in schools of medicine in the United States and Canada. A request was made that specimens of lungs from cadavers used for dissection purposes which were known, or suspected, to have had pulmonary tuberculosis during life be removed under as sterile conditions as possible and sent to us for study. It was requested that specimens be taken from previously undisturbed portions of the diseased lung; that they be removed under as aseptic conditions as possible, using clean sterilized instruments. It was also requested that specimens should be large enough that untouched areas could be dissected out upon receipt. Specimens were to be mailed in clean, preferably sterile, jars or other glass containers. On each specimen submitted, certain pertinent information was requested, to include the date of death, the stated cause of death, the date of embalming, the type and strength of embalming fluid used, the method of storage after embalming and the date the specimen was taken from the cadaver.

Splendid cooperation was experienced and, in all, specimens were received from a total of sixteen different sources.

Upon receipt the specimens were placed in refrigeration until examination and use of them could be made. Each specimen was separately examined and dissected with clean, sterilized instruments for gross evidence of tuberculosis. A number of specimens were discarded because in the portion submitted to us no gross evidence of tuberculosis could be found. From those specimens showing gross evidence of tuberculosis pieces of

[1] From the Trudeau Sanatorium and Edward L. Trudeau Foundation.

involved tissue were removed, crushed and smeared, stained by Ziehl-Neelsen method, and examined at once for presence of tubercle bacilli. No tissue was used in the experiment, with one exception which did not show tubercle bacilli on direct smear. Portions of suspected tissue were also taken for histological study. After direct smear examination, portions of tissue from the same area of the specimen were triturated in physiological saline and injected into guinea pigs previously found negative to 5 per cent Old Tuberculin. Two guinea pigs were used for each specimen. Following the inoculation, tuberculin tests were performed at the end of two and three months. The usual criterion of at least 5 mm. of induration with or without erythema was used in reading the tuberculin tests. Three months after inoculation all surviving guinea pigs were sacrificed and examined for evidence of tuberculosis.

RESULTS

Interval between death and embalming: In the 24 specimens used in the experiment, the interval between death and embalming varied from 0 to 15 days with an average interval of slightly over 4 days. During this interval the bodies were presumably refrigerated the greater part of the time.

Method of embalming: All of the bodies were embalmed by the departments of anatomy to which they were sent, but in a few instances there was primary embalming of a commercial type by an undertaker or professional embalmer followed by a second embalming after delivery to the school of medicine. The method of the primary embalming is unknown.

The embalming methods used varied somewhat in detail as did the fluids. There was no one ingredient common to all the fluids. Phenol in concentrations of from 4.5 to 33 per cent was the most common constituent. Then followed glycerin in concentrations of 16 to 40 per cent; alcohol 10 to 45 per cent; and lastly formalin 3 to 25 per cent. The most commonly used combination contained all four substances. Some of the fluids contained, besides water, such other materials in small amounts as sodium chloride, sopronol, potassium nitrate, and arsenic trioxide. So far as could be seen in this rather limited amount of material, all types of fluid seemed to preserve the tissues equally well.

Duration of embalming: A factor which might have a definite influence on the survival of the tubercle bacilli is the length of time between death of the host and the attempt at isolation in a new, living host; in short, the interval between death of the bodies in question and the subsequent inoculation into guinea pigs. The ultimate length of life of tubercle bacilli in an unembalmed, refrigerated body is not known. The writers previously found in one body that tubercle bacilli remained viable in large numbers under such conditions for at least three months. Thus, the fate of the organisms is possibly the resultant of two factors, namely, the effect of death of the host, and of the embalming fluid. There was not sufficient variety of material to make a comparison of the various time intervals after death with each of the various fluids. However, in the 24 specimens the interval between death of the host and injection of the bacilli-laden tissues into guinea pigs varied from three months to four years. The average was twenty-two and one-half months. The body which had been embalmed for only three months contained many tubercle bacilli on direct smear of the tissue. The embalming fluid used consisted of 25 per cent glycerin, 25 per cent formaldehyde and 50 per

cent water. Four bodies had been embalmed only six months; all showed many organisms on direct smear. One had been embalmed with the 25 per cent glycerin, 25 per cent formaldehyde, 50 per cent water formula; one with arsenic trioxide, phenol, formalin, water and KNO_3; and one with a formula the composition of which was uncertain.

Identification of organisms direct from tissues: In all instances material, usually caseous, was smeared directly on glass slides, stained for acid-fast organisms in the usual manner and examined at once before guinea pig inoculation. An approximation of the number of organisms seen was made, using the terms *many, few, occasional* and *none*. Thirteen of the 24 specimens were rated as having many organisms (usually this meant that the slide showed a myriad of organisms); four were rated as showing few organisms; six were rated as showing occasional; and there was only one specimen which showed no organisms on smear. In the case of this last named specimen, however, the stated cause of death was pulmonary tuberculosis and there were both gross and histologic evidences of tuberculosis. On smear the organisms from all specimens were typical acid-fast organisms which resembled tubercle bacilli in every respect and presented no abnormalities of staining properties or morphology.

Histologic examination of tissues: The tissue taken from the specimens for histological study in every instance showed typical tuberculous reactions.

Results of tuberculin tests: All guinea pigs were tuberculin skin tested two and three months after inoculation. Tests were made with 5 per cent Old Tuberculin. Table 1 shows the results of these testings. The usual standard for a positive reading of 5 by 5 mm. of induration with or without erythema was used. A 5 to 10 mm. reaction was rated as 1+; 10 to 20 mm. as 2+. The guinea pigs from 17 of the 24 specimens were entirely negative throughout the experiment with respect to tuberculin reactions. Some of the test animals showed definitely positive reactions at the end of two months but showed a negative or doubtful reaction at the end of three months. The animals inoculated from one specimen, A-25, were notable in that both guinea pigs showed definitely positive reactions when examined at two and again at three months after injection. Both of these animals were also the only ones of the entire group which showed any abnormality on postmortem, namely, slight, firm, noncaseating enlargement of the inguinal and iliac lymph nodes with no gross evidence of tubercle formation.

It is not unjustified to speculate that the positive tuberculin reactions with negative evidence of disease were induced by the presence of large numbers of dead tubercle bacilli. The reality of the establishment of hypersensitivity in such a manner has been well established (2, 3). In the present study, the measurement of the numbers of tubercle bacilli present was very rough but possibly gave some comparative estimate. In those 7 animals which at two or three months showed a positive tuberculin reaction, the rating of number of tubercle bacilli on smear was many in 5, occasional in one, and none in one. On the other hand there were 8 specimens rated as having many organisms whose inoculated guinea pigs showed no, or questionable, evidence of a positive reaction.

Results of guinea pig inoculation: In all, 48 guinea pigs were inoculated, two

TABLE I

Data on specimens of embalmed lung tissue, arranged in order of duration of embalming

SPECIMEN NUMBER	INTERVAL DEATH TO EMBALMING	INTERVAL EMBALMING TO ANIMAL INOCULATION	EMBALMING FLUID	METHOD OF STORAGE AFTER EMBALMING AND BEFORE DISSECTION	NUMBER ORGANISMS IN SMEAR	POSTINOCULATION TUBERCULIN TEST			GUINEA PIG AUTOPSY 12 WEEKS AFTER INOCULATION
	days	*months*					2 months	3 months	
A-14	7	3	Formalin—25 per cent Glycerin—25 per cent H_2O—50 per cent	Refrigerated at 32°F.	Many	Pig 1..... Pig 2.....	0 0	0 0	Negative Negative
A-4	15	6	Arsenic trioxide, phenol, formalin, H_2O, KNO_3	Immersed in 4 per cent aqueous phenol at room temperature	Many	Pig 1.....	Not done	Not done	Pig died 28 days after inoculation. No tuberculosis.
A-6	7	6	Same as A-4	Same as A-4	None	Pig 2.....	0	0	Negative
A-9	?	6	Same as A-4 or A-7	Same as A-4	Many	Pig 1..... Pig 2.....	0 1+	0 Not done	Negative Died 50 days after inoculation of streptococcus pneumonia. No tuberculosis.
A-19	5	6	Same as A-14	Same as A-14	Many	Pig 1..... Pig 2.....	0 1+	0 0	Negative Negative
A-16	2	7	Same as A-14	Same as A-14	Many	Pig 1..... Pig 2.....	1+ 0	0 0	Negative Negative
						Pig 1..... Pig 2.....	0 0	0 0	Negative Negative

A-7	10	11	Alcohol—39 per cent Formalin—3 per cent Glycerin—39 per cent Phenol—10 per cent	Same as A-4	Many	Pig 1..... Pig 2.....	1+ Not done	1+ Not done	Negative Died 21 days after inoculation of streptococcus pneumonia. No tuberculosis.
A-15	1	11	Same as A-14	Same as A-14	Occasional	Pig 1..... Pig 2.....	1+ 1+	0 ±	Negative Negative
A-5	7	12	Same as A-7	Same as A-4	Many	Pig 1..... Pig 2.....	0 0	± Not done	Negative Died 45 days after inoculation. No tuberculosis.
A-13	5	13	Same as A-14	Same as A-14	Few	Pig 1..... Pig 2.....	0 0	0 0	Negative Negative
A-18	0	14	Same as A-14	Same as A-14	Many	Pig 1..... Pig 2.....	Not done 0	Not done 0	Died accidental death 18 days after inoculation. No tuberculosis. Negative
A-17	4	16	Same as A-14	Same as A-14	Few	Pig 1..... Pig 2.....	0 0	0 Not done	Negative Died 67 days after inoculation of streptococcus pneumonia. No tuberculosis.
A-30	3	18	Alcohol—33 per cent Glycerin—33 per cent Phenol—33 per cent	Immersed in 2 per cent phenol	Occasional	Pig 1..... Pig 2.....	0 0	0 0	Negative Negative

TABLE I—*Continued*

SPECIMEN NUMBER	INTERVAL DEATH TO EMBALMING	INTERVAL EMBALMING TO ANIMAL INOCULATION	EMBALMING FLUID	METHOD OF STORAGE AFTER EMBALMING AND BEFORE DISSECTION	NUMBER ORGANISMS IN SMEAR	POSTINOCULATION TUBERCULIN TEST			GUINEA PIG AUTOPSY 12 WEEKS AFTER INOCULATION
	days	*months*					2 months	3 months	
A-11	5	26	95% Alcohol—33 per cent Glycerin—33 per cent Phenol—33 per cent	Immersed in 5 per cent cresylic acid solution	Many	Pig 1..... Pig 2.....	0 0	0 0	Negative Negative
A-12	4	28	Same as A-11	Same as A-11	Many	Pig 1..... Pig 2.....	2+ 1+	± ±	Negative Negative
A-10	4	29	Same as A-11	Same as A-11	Many	Pig 1..... Pig 2.....	0 ±	0 0	Negative Negative
A-1	5	36	95% Alcohol—33 per cent Formalin—9 per cent Glycerin—16 per cent NaCl—1.6 per cent Phenol—4.5 per cent Sopronol—5 per cent H_2O—31 per cent	Greased, wrapped, stored at 37°F.	Many	Pig 1..... Pig 2.....	0 0	0 0	Negative Negative
A-23	4	38	Alcohol 10 per cent Formalin Glycerin Phenol	Immersed in 2 to 3 per cent phenol	Occasional	Pig 1..... Pig 2.....	0 0	0 0	Negative Negative

A-24	2	39	Same as A-23	Same as A-23	Few	Pig 1.....	0	0	Negative
						Pig 2.....	±	0	Negative
A-29	2	40	Same as A-30	Same as A-30	Occasional	Pig 1.....	0	0	Negative
						Pig 2.....	0	0	Negative
A-28	1	42	Same as A-23	Same as A-23	Occasional	Pig 1.....	0	0	Negative
						Pig 2.....	0	0	Negative
A-25	1	45	Same as A-23	Same as A-23	Many	Pig 1.....	1+	1+	Sl. enlarged, firm, non-caseous inguinal and iliac nodes. No tuberculosis.
						Pig 2.....	1+	1+	Sl. enlarged, firm, non-caseous inguinal and iliac nodes. No tuberculosis.
A-27	1	46	Same as A-23	Same as A-23	Occasional	Pig 1.....	0	0	Negative
						Pig 2.....	0	0	Negative
A-26	3	48	Same as A-23	Same as A-23	Few	Pig 1.....	0	0	Negative
						Pig 2.....	0	0	Negative

for each of the 24 specimens. During the course of the experiment 6 animals died: one of an accident, 3 of pneumonia, and 2 of unknown cause. None of these animals showed any evidence of tuberculosis at the time of death which occurred from 18 to 67 days after inoculation. In no instance did both the animals inoculated from any one specimen die so that at least one animal inoculated from each specimen was available to complete the experiment.

No animals died of tuberculosis during the experiment. All surviving animals were sacrificed at the end of 12 weeks. No gross evidence of tuberculosis was found in any of them. In both animals of one specimen (A-25) there was slight, firm, noncaseating enlargement of the inguinal and iliac lymph nodes.

COMMENT

In view of the all too frequent infection of medical students with tuberculosis and the observation that some primary infections regularly occur during the first year of the medical curriculum (1), it seemed advisable to establish whether or not the dissection of cadavers which had pulmonary tuberculosis in life constitutes a possible source of infection. The question of viability of tubercle bacilli in embalmed tissues doesnot seem to have been dealt with in the literature on tuberculosis. The present study does not pretend to be a definitive one in that no attempt was made to establish standard embalming techniques nor minimal or maximal survival times. The study simply aims to show whether tubercle bacilli survive in the lungs of anatomy dissection cadavers under the usual techniques and practices of schools of medicine. It would appear from this study that in all probability the organisms are nonviable after three to six months with the methods in general use. No instance was encountered where the body was used for dissection sooner than three months after embalming.

Since this study was confined to the lungs of the embalmed bodies, the findings and conclusions can strictly be applied only to this organ tissue. It is conceivable that the penetration of embalming fluids to other tissues might not be as complete and that there might thus be occasional survival elsewhere. It is also possible that at times a portion of a lung or other organ might not be adequately embalmed because of blockage of embalming fluid from that area by an occluded vessel. In such an instance it is conceivable that there might be local survival of tubercle bacilli.

The development of positive tuberculin reactions in some of the animals without evidence of disease suggests the possibility that tuberculous dissection material might be a source of a tuberculin conversion because of the presence of dead tubercle bacilli if a sufficient amount of material entered the body. Such a possibility, however, seems quite remote and hardly worth consideration.

SUMMARY AND CONCLUSIONS

1. Tubercle bacillus laden lung tissue from 24 anatomy dissection cadavers obtained from schools of medicine was injected into guinea pigs.

2. Some of the injected animals developed positive tuberculin tests at the end of two or three months.

3. No guinea pigs died of tuberculosis and none showed evidence of disease at autopsy three months after inoculation.

4. It is concluded that under the conditions ordinarily prevailing tubercle bacilli are not viable in lungs of embalmed Medical School dissection cadavers at the time of dissection.

SUMARIO Y CONCLUSIONES

Viabilidad de los Bacilos Tuberculosos en el Tejido Pulmonar Humano Embalsamado

1. Inyectóse en cobayos tejido pulmonar cargado con bacilos tuberculosos, obtenido de 24 cadáveres utilizados para disección anatómica en facultades de medicina.

2. Algunos de los animales inyectados manifestaron reacciones positivas a la tuberculina al cabo de dos o tres meses.

3. Ningún cobayo murió de tuberculosis ni ninguno mostró signos de enfermedad en la autopsia a los tres meses de la inoculación.

4. Dedúcese que, en las condiciones que reinan habitualmente, los bacilos tuberculosos no son viables en los pulmones de los cadáveres embalsamados para disección al hacerse la última.

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ACQUIRED RESISTANCE OF M. TUBERCULOSIS TO STREPTOMYCIN IN VITRO AND ITS BEHAVIOR IN EXPERIMENTAL INFECTIONS[1, 2]

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The development of resistance *in vivo* of the tubercle bacillus to streptomycin in man in the course of treatment was reported by Youmans and associates (1). More recently Williston and Youmans (2) have also shown that the degree of acquired resistance *in vitro* varied with different strains. Using a culture medium consisting of a synthetic broth with plasma to which streptomycin was added, 4 out of 18 strains failed to acquire resistance, one strain developed a twofold resistance, 3 only a fourfold resistance while 9 strains showed a thousandfold resistance. These results are in accord with those of Karlson, Feldman and Hinshaw (3) who demonstrated that resistance of strains from treated tuberculous patients varied from .08 to 2,000 γ per cc. and that the acquired resistance was not lost upon passage through guinea pigs and subsequently maintained for many weeks on solid culture media.

The present report presents data on the acquisition of resistance *in vitro* by the virulent slow growing human strain A27; the difference in growth rate of the resistant and control strains; the relative degree of invasiveness of these strains in the chorio-allantois of the chick embryo and in the guinea pig; and the efficacy of streptomycin therapy in experimental infections in guinea pigs inoculated with the resistant strain.

RESULTS

Acquired resistance in vitro and the growth rate of the resistant strain: The human strain A27 used in these experiments had initially a sensitivity of 1.0 to 1.5 γ per cc. (4). After a series of seventeen successive passages in Kirchner's medium with increasing concentrations of streptomycin, beginning with an initial concentration of 0.5 γ per cc., the strain grew well in a concentration of 5.0 γ per cc.

After the strain had developed a tolerance to 4.0 γ per cc. streptomycin, it was transferred to the control medium for two generations totaling approximately 70 days and then its growth rate was compared with that of the parent strain. Three experiments were set up, each in triplicate. The average growth of the 9 flasks for both the control and resistant strains was determined at intervals of 5 days and recorded as previously described (5).

At the end of 45 days, when the experiment was terminated, the growth of the control strain covered the whole surface of the culture medium and was rated 4.0,

[1] From the Experimental Biology and Medicine Institute, National Institute of Health, Bethesda, Maryland.

[2] Presented at the Antibiotics Study Section Seminar, National Institute of Health, October 1, 1947.

[3] Biostatistician, Division of Public Health Methods, U. S. Public Health Service.

while the growth of the streptomycin-resistant strain averaged 2.2. After the initial lag period of approximately 12 days, the growth curve of the control strain was found to be consistently higher than that of the resistant strain (figure 1) and at the end of the experimental period of 45 days the growth of the resistant strain was about 55 per cent of that of the control strain. Since the rate of growth of the two strains within a given period of time may be the basis for testing tuberculostatic action *in vitro*, it was logical to assume that the comparison of the growth curves over the entire experimental period would furnish more information than the single comparison of growth at the termination of the experiment. In this

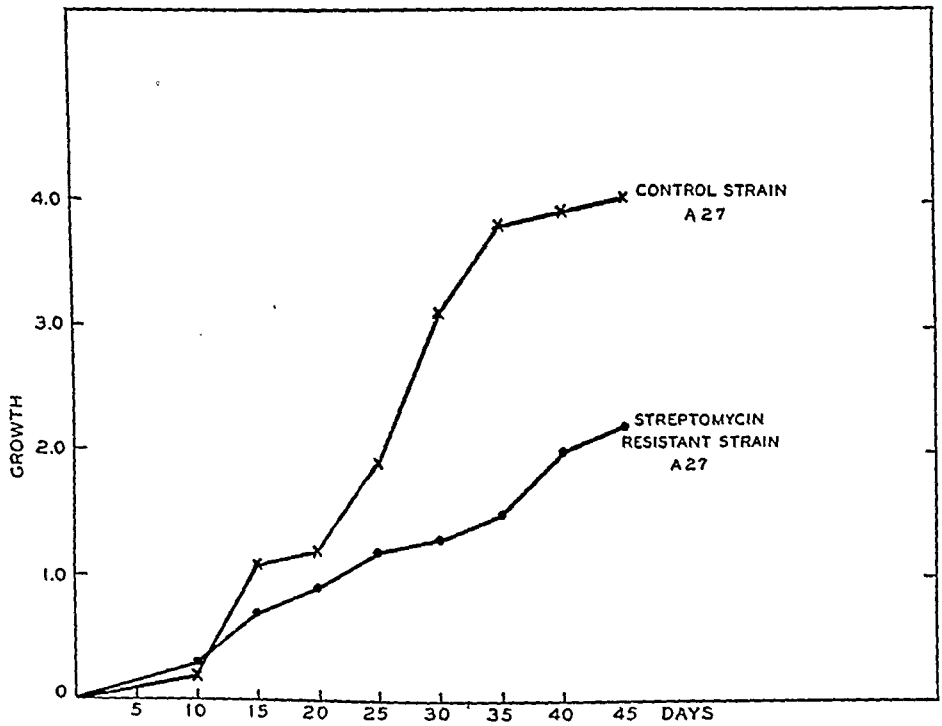

FIG. 1. Difference in growth rate of streptomycin-resistant strain (A27) as compared with the control strain in Kirchner's medium. The points represent the average growth of 9 flasks at successive intervals of time.

respect the problem is similar to that of a bio-assay (6, 7) in which the effect of a biological product is determined over a range of concentrations and not at just one concentration.

In adopting a bio-assay technique it was necessary to make the assumption that the logarithm of the growth measurement has a linear relation to the time. Tests of this assumption have been made on the data under consideration and no significant departure from linearity has been found. In the statistical analysis of the difference in the growth of the control and resistant strains, the mean growth readings for each strain were converted into logarithms and straight lines fitted to the points by the method of least squares (8). In figure 2 are represented the recti-

fied growth curves shown in figure 1. The equations of these rectified curves as determined by least squares are—

$$y_1 = 0.0054 + 0.015\ t_1$$

and

$$y_2 = 0.3305 + 0.016\ t_2$$

where y_1 and t_1 are respectively the log growth and the time for the control strain, and y_2 and t_2 stand for the same variables in the resistant strain.

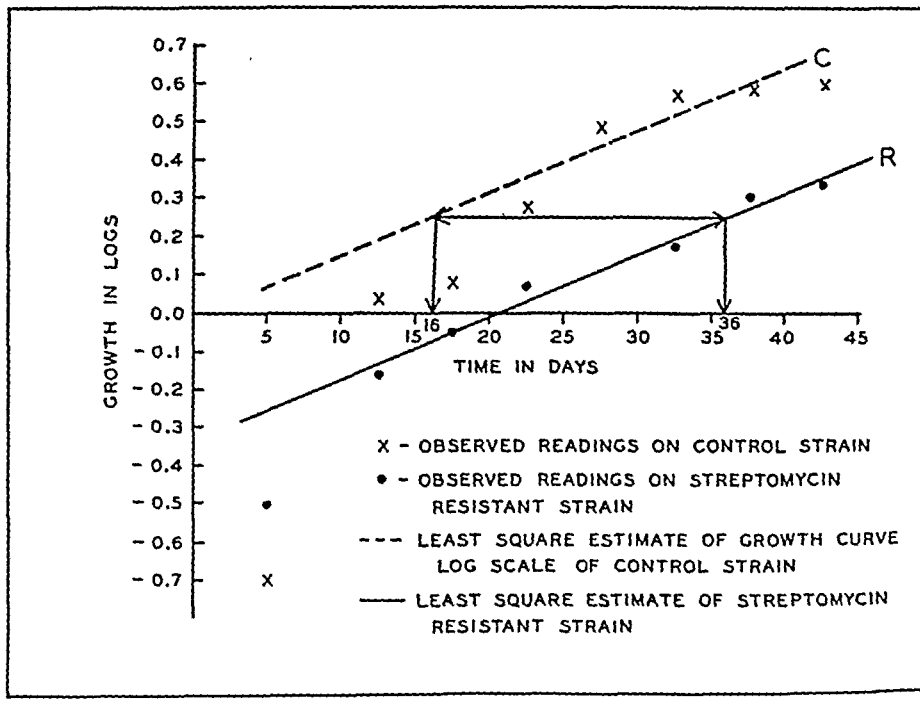

FIG. 2. Log growth of control and streptomycin-resistant strains with adjusted curves by the method of least squares. The difference of 36 and 16, or 20, days represents the difference in time required for the resistant strain to reach the same amount of growth as the control strain.

These rectified curves are also plotted in figure 3 and 4 on semi-log paper. In these figures are shown the curves representing a multiple of the standard error of estimation of the average growth at each point along the curve. In figure 3 this is represented for the control strain by the 95 per cent confidence band.[4] The minimum error of estimation occurs near the growth of 1.8 which in log units is near 0.25 (figure 2). In figure 4 the confidence band for the resistant strain is shown.

[4] Since the growth rate was greater in the control strain, the observable deviations from the mean were greater. This is represented by the wider spread of the confidence band (figure 3) as compared with the confidence band of the resistant strain (figure 4). This also explains the deviation of some of the points from the straight line C in figure 2.

Using the log growth of 0.25 as the estimated growth at the point of approximate minimum error, the time when the control and resistant strains reached this growth could then be determined. In figure 2 this point on the curve of the control strain corresponds to 16 days. The same amount of growth on the curve of the resistant strain corresponds to 36 days. The difference of 36 minus 16, or 20 days, is the measure of retardation of growth of the resistant strain.

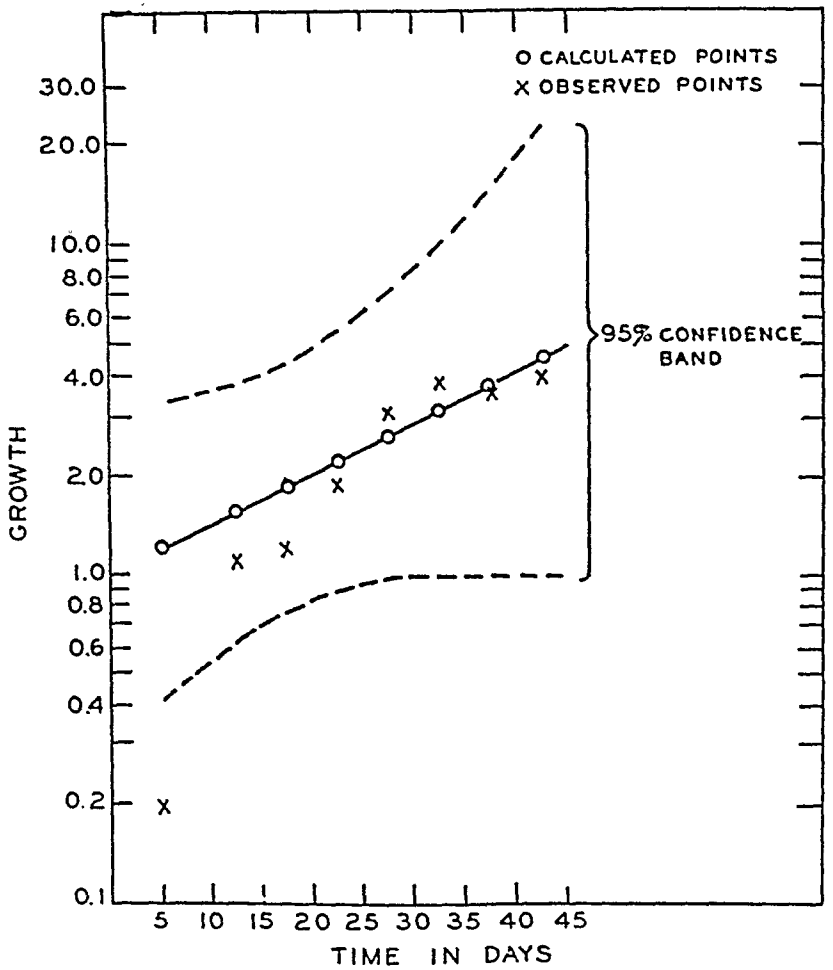

FIG. 3. Rectified growth curve of the control strain and the 95 per cent confidence band. The minimum error of estimation is the point on the growth band which most closely approaches the confidence band.

Invasiveness of the streptomycin-resistant and control strains in the chorio-allantoic membrane of the chick embryo: In order to determine the relative degree of invasiveness of the 2 strains in the chorio-allantoic membrane of the chick embryo, 7 experiments were carried out in which the resistant and the control strains were each inoculated on the membranes of the 8 day old chick embryo. The technique of preparation and mode of inoculation were the same as previously described (9, 5) with 1.0 mg. of bacilli in each 0.2 cc. of inoculum. In these experiments the

resistant strain was obtained from the fifth to the sixteenth passage in 0.5, 1.0 and 4.0 γ per cc. of streptomycin respectively. The membranes were harvested 6 days after inoculation and the inoculated areas fixed, excised, and the size and number of tubercles counted. Table 1 gives the average tubercles per membrane for the experimental and control groups for the 7 experiments. From the data obtained during the brief 6 day experimental period, it was not possible to demonstrate a consistent difference in the degree of invasiveness of the resistant and control strains. However, some of the same suspensions used for the chick embryo tests

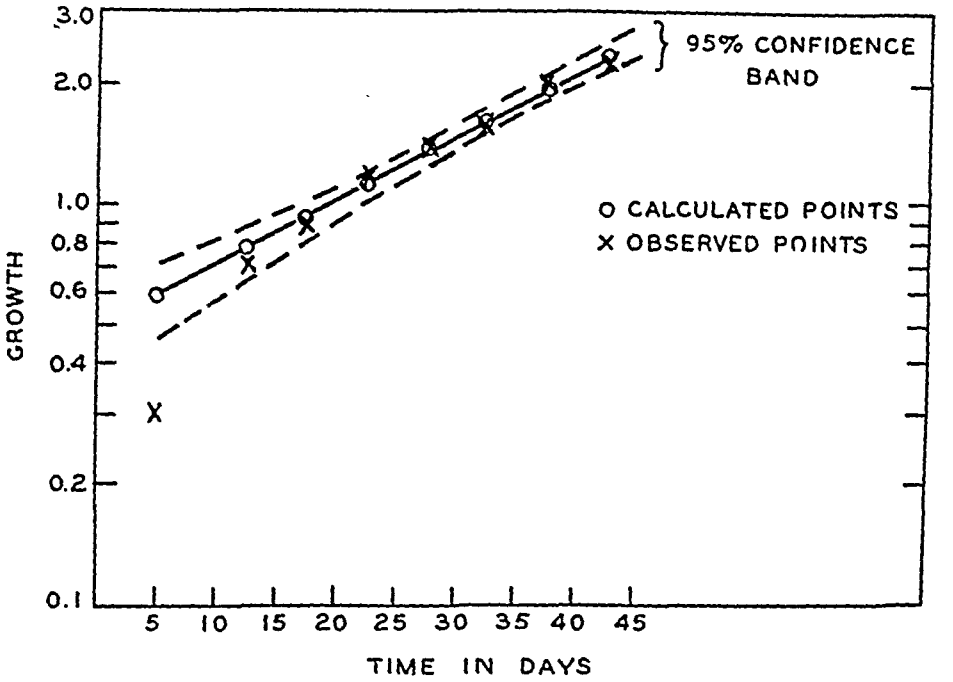

FIG. 4. Rectified growth curve of the slowly growing resistant strain with the 95 per cent confidence band. The curves representing the limits of the confidence band are based upon a multiple of the standard error of estimation of the average growth at each point along the growth curve.

were also used for inoculation into guinea pigs so that the course of the infection might be observed during a longer period.

Pathogenicity of the resistant and control strains after inoculation into guinea pigs: To determine the degree of invasiveness of the streptomycin-resistant strain as compared with the control in guinea pigs, 4 experiments were carried out with some of the same bacterial suspensions as were used in the preceding experiments. In experiment 1, inoculations were made from suspensions after the fifth passage; in experiment 2 after the seventh passage in 0.5 γ per cc. streptomycin; in experiment 3 from suspensions of the twelfth transfer in 3.5 γ per cc. streptomycin; and in experiment 4 from suspensions of cultures of the sixteenth transfer in 4.0 γ per cc. streptomycin. At the time of the twelfth passage the strain in strepto-

mycin had developed a two-to threefold resistance. The dosages injected intraperitoneally were as follows: experiments 1 and 2, 1 mg.; experiment 3, 0.25 mg.; and experiment 4, 0.5 mg.

Experiments 1, 2 and 3 were terminated 42 and 43 days after inoculation while experiment 4 was terminated 105 and 106 days after inoculation. The degree of invasiveness of the strain was based upon the extent of tuberculous involvement of the liver, spleen, omentum, peritoneum and kidneys and lungs as previously described (10). In experiment 1, the difference in the tuberculosis index between the control and experimental groups was negligible. In the last 3 experiments, the tuberculosis index was slightly less in the experimental groups than in the control groups (table 2).

TABLE 1

Invasiveness of the streptomycin-resistant (R) and control (C) strains of A27 in the chorio-allantoic membrane of the chick embryo

EXPERIMENT NUMBER	STRAIN OF TUBERCLE BACILLI*	NUMBER OF PASSAGES AND AMOUNTS OF STREPTOMYCIN	NUMBER OF EMBRYOS SURVIVING	AVERAGE NUMBER OF TUBERCLES PER MEMBRANE			TOTAL AVERAGE NUMBER OF TUBERCLES PER MEMBRANE
		γ per cc.		Large	Medium	Small	
257	R	Fifth—0.5	16	0.7	3.1	14.0	17
	C		16	1.2	3.8	13.1	17
264	R	Sixth—0.5	8	0.2	1.1	6.7	8
	C		10	0.5	1.5	12.2	14
268	R	Seventh—0.5	10	1.0	3.3	5.6	10
	C		12	0.7	1.0	6.3	8
282	R	Tenth—1.0	13	0.0	0.2	17.0	17
	C		13	0.1	0.2	17.0	18
300	R	Fourteenth—4.0	22	0.2	0.3	0.9	1.3
	C		20	0.2	0.1	0.8	1.0
301	R	Fifteenth—4.0	23	0.2	0.3	4.0	4.6
	C		21	0.4	0.9	9.6	10.9
302	R	Sixteenth—4.0	25	0.2	0.6	1.4	2.1
	C		18	0.2	0.5	1.2	1.4

* R = resistant; C = control.

All the inoculated guinea pigs were weighed once a week and the average weight for each group calculated and plotted. Inspection of figures 5 and 6 will show that in each of these experiments the weight curves of the guinea pigs inoculated with the resistant strain were consistently higher than those inoculated with the control strain.

The differences in the weight curves and the tuberculosis index of the guinea pigs inoculated with the streptomycin-resistant strain and the control strain may possibly be explained by the difference in growth rates of the 2 strains as shown in the *in vitro* experiments or by a possible reduction in virulence or both. Yegian, Budd and Middlebrook (11) have demonstrated a slower growth rate of *M. ranae* with acquired resistance to certain sulfonamides.

Streptomycin therapy in guinea pigs inoculated with streptomycin-resistant and

control strains: In order to determine the effect of streptomycin therapy in guinea pigs inoculated with the streptomycin-resistant strain, two groups of 40 guinea pigs each were inoculated intraperitoneally with 0.5 mg. of tubercle bacilli; one group received the control strain A27 and the other received the streptomycin-resistant strain. These groups were subdivided into 2 groups of 20 each, one of which served as control and the other was treated with streptomycin intramuscularly for a period of 12 weeks. Treatment was begun the day after infection. The drug was administered in 10 mg. per Kg. doses twice daily 5 days a week with a double dose on the fifth day. At 13 weeks the survivors were tuberculin tested with .01 mg. PPD intracutaneously. The experiment was terminated at 97 to

TABLE 2

Pathogenicity of streptomycin-resistant strains A27 in guinea pigs

EXPERIMENT NUMBER	TUBERCULOSIS STRAIN A27	NUMBER OF TRANSFERS IN STREPTOMYCIN	NUMBER OF GUINEA PIGS INOCULATED	DOSE INOCULATED INTRAPERITONEALLY	NUMBER OF ANIMALS DIED BEFORE TERMINATION OF EXPERIMENT	INTERVAL BETWEEN INOCULATION AND AUTOPSY	AVERAGE TUBERCULOSIS INDEX FOR THE GROUP
				mg.		*days*	
1	Streptomycin-resistant	Fifth transfer in 0.5 γ per cc.	17	1.0	1	42	10
	Control	—	17	1.0	1	42	11
2	Streptomycin-resistant	Seventh transfer in 0.5 γ per cc.	18	1.0	5	43	10
	Control	—	18	1.0	4	43	16
3	Streptomycin-resistant	Twelfth transfer, last 3 in 3.5 γ per cc.	19	0.25	0	42	8
	Control	—	18	0.25	1	42	10
4	Streptomycin-resistant	Sixteenth transfer, last 4 in 4.0 γ per cc.	20	0.5	12	97 to 107	9
	Control	—	20	0.5	16		12

106 days after inoculation, when the survivors were killed and the extent of tuberculous involvement noted and rated as previously described (10).

In figure 6 are shown the weight curves of the 4 groups of guinea pigs. As previously pointed out, the weight curve of the untreated animals which were inoculated with the resistant strain (curve R) was consistently higher than that of the untreated animals inoculated with the control strain (curve C). The weight curve of the group infected with the control strain and treated with streptomycin (curve CT) was the highest of the 4 groups. At 11 weeks the average weight of the animals of this group was 537 Gm. while that of the untreated was 363 Gm. Both groups infected with the resistant strain, whether treated or not treated with

streptomycin, had a somewhat higher weight curve than the untreated group infected with the control strain.

The slight drop in weight at 13 weeks was probably a result of the intracutaneous administration of .01 mg. of PPD. Of the 10 animals given PPD in the un-

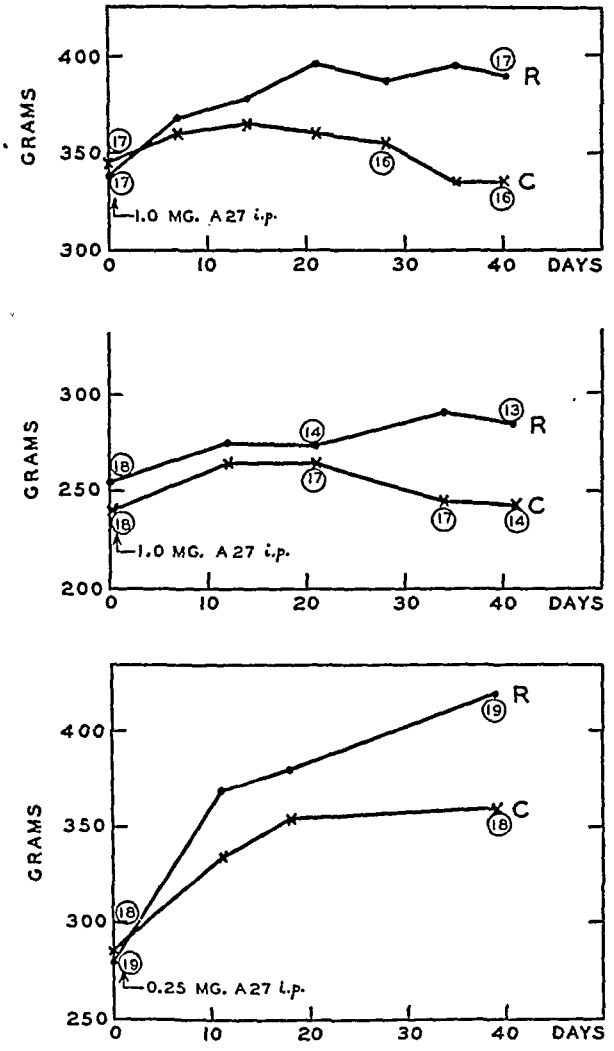

FIG. 5. Average weight curves of guinea pigs inoculated with the streptomycin-resistant strain (A27-R) and the control strain (A27-C):

Experiment 1. (Top) After 5 transfers in 0.5 γ per cc. streptomycin.

Experiment 2. (Center) After 7 transfers in 0.5 γ per cc. streptomycin.

Experiment 3. (Bottom) After 12 transfers in 0.5 γ to 3.5 γ per cc. streptomycin (Table 2).

treated group inoculated with the resistant strain, 7 were positive; while of the 10 of the treated group, 8 gave a pronounced positive reaction. In the untreated control group, 4 of the 5 survivors gave positive reactions while all of the 17 in the treated group, which had received the control A27 strain, gave positive PPD reactions.

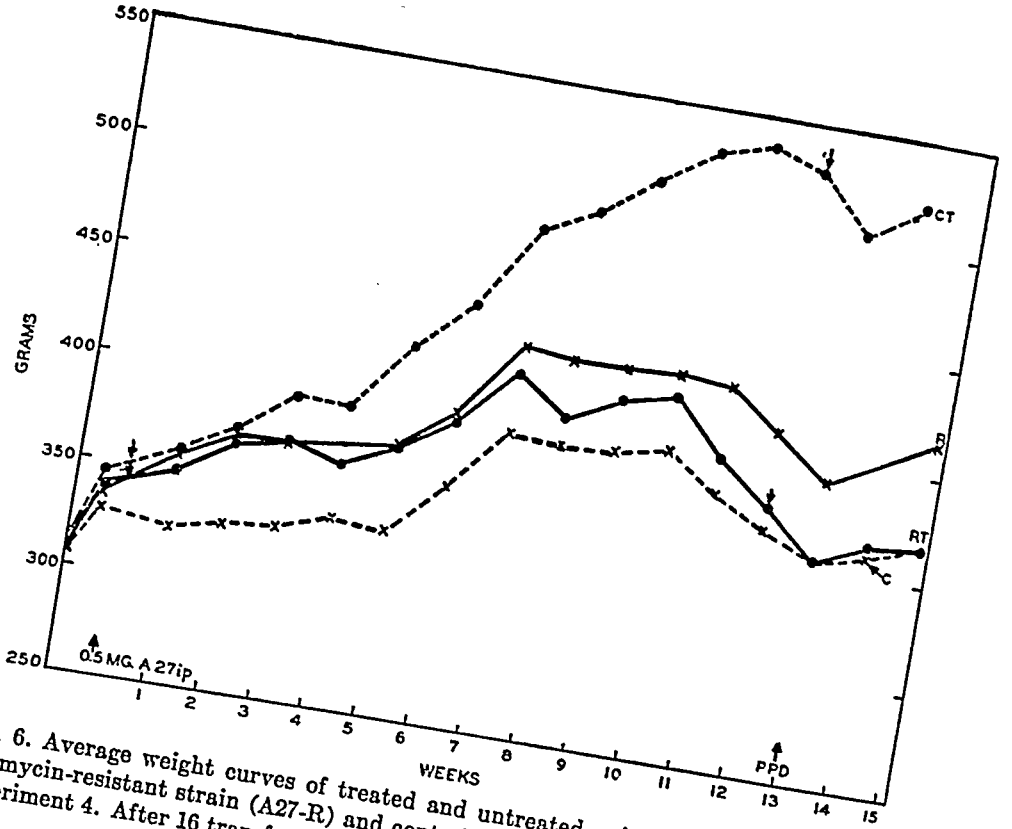

FIG. 6. Average weight curves of treated and untreated guinea pigs inoculated with streptomycin-resistant strain (A27-R) and control strain (A27-C).
Experiment 4. After 16 transfers in 0.5 to 4.0 γ per cc. streptomycin (Tables 2, 3).

TABLE 3

Virulence of streptomycin-resistant tubercle bacilli in guinea pigs and effect of treatment with streptomycin

GUINEA PIG NUMBER	CONTROL STRAIN A27				STREPTOMYCIN-RESISTANT STRAIN A27			
	Control		Treated		Control		Treated	
	Survived days	Tuberculosis	Survived days	Tuberculosis	Survived days	Tuberculosis	Survived days	Tuberculosis
1	47	10	98	1	K* 106	14	91	13
2	79	16	97	2	52	10	K 105	15
3	K 105	4	97	1	K 106	11	K 106	13
4	90	12	98	1	75	17	K 106	11
5	74	12	98	2	K 106	6	58	5
6	K 105	7	98	2	K 105	14	K 106	12
7	93	12	97	0‡	89	11	64	18
8	67	14	98	1	58	9	91	7
9	76	7	98	4	105	12	84	11
10	84	13	97	1	58	6	88	16
11	62	20	D† 33	3	K 106	5	61	6
12	81	12	D 89	4	90	14	95	17
13	74	11	98	2	89	15	36	10
14	K 105	5	D 12	0	K 106	4	K 106	8
15	75	11	97	3	K 106	4	K 106	19
16	K 105	15	98	1	84	9	90	10
17	74	18	98	5	K 105	3	K 106	11
18	37	15	98	4	105	6	92	12
19	83	19	97	3	83	9	K 105	16
20	77	11	98	2	58	5	89	14
Mortality per cent	80		15		60		60	
Average tuberculosis index		12.2		2.2		9.2		12.2

* K—killed, † D—died, ‡ subculture of spleen negative after 35 days.

The survival time and the extent of tuberculous involvement for each animal of the 4 groups are given in table 3. At the termination of the experiment the mortality of the control group was 80 per cent while that of the group infected with the control strain and treated with streptomycin was 15 per cent. The mortality for both the streptomycin treated and untreated groups of guinea pigs which had been inoculated with the resistant strain was 60 per cent. The tuberculosis index of the guinea pigs inoculated with the A27 control strain was 12.2 while that of the control strain treated with streptomycin was 2.2. There was no difference in the tuberculosis index between the untreated group infected with the control strain and the treated group infected with the resistant strain. The untreated group infected with the resistant strain had a tuberculosis index of 9.2 while that of the treated group infected with the resistant strain was 12.2. The results of this experiment clearly indicate that treatment with streptomycin of tuberculous guinea pigs infected with the streptomycin-resistant strain of tubercle bacilli had no therapeutic effect on the course of the disease.

SUMMARY

1. The slow growing virulent human strain of tubercle bacilli (A27) has been made resistant to streptomycin by successive passages in Kirchner's medium with increasing concentrations of streptomycin. After 17 passages the degree of induced resistance was from an original sensitivity of 1.0 to 1.5 γ per cc. to a tolerance of 5.0 γ per cc.

2. The growth of the resistant strain in Kirchner's medium was slower when compared with the control strain. When the growth curves were subjected to statistical analysis it was found that the growth of the resistant strain was retarded on an average by 20 days.

3. The virulence of the resistant strain was indistinguishable from that of the control strain when tested by the short term technique of inoculation of the chorio-allantois of the chick embryo.

4. When tested in guinea pigs by intraperitoneal inoculation of 0.25 mg. to 1.0 mg. of tubercle bacilli and terminating the experiments 42 days after infection, the results of three tests showed a slightly reduced tuberculosis index and a higher average weight curve for the animals infected with the resistant strain. In a fourth experiment with 0.5 mg. of tubercle bacilli as the infecting dose with each strain, and the experiment terminated 14 to 15 weeks after infection, the mortality for the group infected with the resistant strain was 60 per cent as compared with 80 per cent for the control group and the tuberculosis index was 9.2 as compared with 12.2 for the controls.

5. Streptomycin therapy in doses which gave a high degree of protection in animals infected with the control strain was ineffective in infections with the resistant strain.

SUMARIO

Resistencia Adquirida del M. Tuberculosis a la Estreptomicina in vitro y Comportamiento del Mismo en las Infecciones Experimentales

1. La cepa humana de bacilos tuberculosos, virulenta y de desarrollo lento (A27) se convirtió en estreptomicinorresistente mediante pases sucesivos por el

medio de Kirchner con concentraciones crecientes de estreptomicina. Tras 17 pases, la intensidad de la resistencia inducida había pasado de la primitiva sensibilidad de 1.0 to 1.5 γ por cc. a una tolerancia de 5 γ por cc.

2. El crecimiento de la cepa resistente en el medio de Kirchner fue más lento que el de la cepa testigo. Al someter las curvas de crecimiento al análisis estadístico, observóse que el retardo del crecimiento de la cepa resistente promediaba veinte días.

3. La virulencia de la cepa resistente era indistinguible de la de la cepa testigo al comprobarla con la técnica breve de inoculación de la corioalantoides del embrión de pollo.

4. Al ensayar en los cobayos la inoculación intraperitoneal de 0.25 a 1.0 mg. de bacilos tuberculosos y terminar el experimento a los cuarenta y dos días de la infección, el resultado de tres ensayos reveló un índice tuberculoso algo menor y una curva ponderal media mayor para los animales infectados con la cepa resistente. En el cuarto experimento con 0.5 mg. de bacilos tuberculosos como dosis infectante con cada cepa, que terminó de catorce a quince semanas después de la infección, la mortalidad para el grupo infectado con la cepa resistente fué de 60 por ciento, comparado con 80 por ciento para el grupo testigo, siendo el índice tuberculoso de 9.2 y 12.2, respectivamente.

5. La estreptomicinoterapia, a dosis que suministraron una protección alta en los animales infectados con la cepa testigo, resultó ineficaz en las infecciones con la cepa resistente.

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ROUTINE EXAMINATION OF SPUTUM FOR ACID-FAST BACILLI[1]

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For the past several years the sputum and gastric contents of all patients admitted to this institution have been routinely examined by both microscopic and cultural methods. In an effort to determine the value of the methods used and the efficacy of the routine as a whole, an analysis has been made of the results obtained over a period of four years from January 1, 1940, when the routine was well established, to January 1, 1944. During this period, very few changes were made in the routine method, technical staff, or type of patient admitted. It has been possible, therefore, to make a fairly reliable evaluation of the relative performance of the various methods over a significant interval.

MATERIALS AND METHODS

Bacteriologic Procedure

The method of bacteriologic examination used routinely in this laboratory is as follows. Upon admission of the patient, three successive sputum specimens are sent to the laboratory as quickly as they can be collected in adequate amount, preferably within a period of ten days. These are examined for acid-fast bacilli by direct smear and, if negative, are concentrated. If all three specimens are negative for acid-fast bacilli, the third specimen is routinely cultured. The second or even the first specimen may occasionally be cultured, however, if the clinical condition of the patient warrants. Following the third admission specimen, the sputum is examined every one or two months, or more often if necessary. If the concentrate is persistently negative for tubercle bacilli, it is routinely cultured. If the sputum is repeatedly negative for tubercle bacilli, or if little or no sputum is available, the gastric contents are examined and all such specimens are cultured. Guinea pig inoculation is used only in special instances, usually in cases in which the roentgenographic appearance of the lesion seems to belie the persistently negative bacteriologic findings.

All smears for microscopic examination are stained individually by the Ziehl-Neelsen method and are examined intensively for a period of not less than five minutes. Concentrates are made by the addition of an equal volume of 3 per cent sodium hydroxide to the sputum or gastric specimen and incubating with frequent shaking for one to two hours. Cultures are made on a minimum of four tubes of the modification of Petragagni's medium described in Diagnostic Standards[3]. About 0.5 cc. of neutralized digest are inoculated into each tube by means of a capillary pipette and the tubes are incubated for a period of not less than two months before being discarded. All positive cultures are smeared for confirmation.

Composition of Material Subjected to Analysis

In the evaluation of the results of this analysis, several points must be borne in mind. The patients admitted to this institution are a rather highly selected

[1] From the Montefiore Hospital Country Sanatorium, Bedford Hills, New York.

[2] Doctor Pinner died on January 7, 1948.

[3] Diagnostic Standards and Classification of Tuberculosis, published by the National Tuberculosis Association.

group and include a large proportion of minimal and moderately advanced infections with a favorable prognosis. A significant number represent cases discovered on mass survey or on army induction examinations. Patients in the terminal stages of the disease are either not admitted or are quickly transferred. This practice is reflected in the very low number of sanatorium deaths, which average only one or two per year and often result from causes other than tuberculosis. It is not possible, therefore, to extend these results except in a very general way to institutions where all types of patients are freely admitted, or to the average office practice.

It must also be borne in mind that the results obtained in evaluating the various methods of examination represent not the actual efficiency of the method but rather its general contribution within the routine procedure described. Each patient received an unspecified number of sputum and gastric examinations conditioned by his physical status, therapeutic regimen, and length of stay, and no single specimen was subjected to all methods of examination. In actual practice, the selective methods routinely employed tend to weight the figures in favor of the microscopic methods. For example, sputum specimens found positive for acid-fast bacilli on direct smear tend to be repeatedly re-examined by this method and are rarely cultured, whereas specimens repeatedly negative are usually concentrated and cultured immediately without prior resort to smear. Furthermore, whereas all cultured sputum specimens are first concentrated and examined microscopically, only 31.9 per cent of concentrated sputum specimens are subsequently cultured. Since the uncultured specimens are those in which the presence of tubercle bacilli can most readily be expected on the basis of past performance and the clinical and roentgenographic appearance of the lesion, in other words, those in which a relatively high percentage of positive cultures is likely to be obtained, the microscopic method is thus favored. In the case of the gastric specimens a more reliable relationship obtains, since practically all specimens are subjected to both concentration and culture.

Results of the Analysis

The performance of the various methods of examination is shown in table 1. In spite of the fact that the microscopic methods are favored, the important contribution of the cultural method is evident. In the case of sputum specimens, culture is still nearly twice as effective in detecting tubercle bacilli as is microscopic examination of the concentrate; in the case of gastric specimens, culture is five times as effective. The significance of these figures is indicated by the relative constancy of these relationships for the four yearly intervals.

The value of the cultural method as an aid in bacteriologic examination for acid-fast bacilli is even more clearly shown in table 2. Of nearly 5,000 sputum and gastric specimens found to be negative for acid-fast bacilli by microscopic examination, an increment of 30 per cent was positive on culture. Furthermore, the remarkably small variation of this percentage when calculated for the various six month intervals is a significant commentary, both on the constancy of performance of technical personnel and culture medium and on the relatively

TABLE 1

Results of the various methods of examination for acid-fast bacilli

YEAR	SPUTUM										GASTRIC						
	Smear			Concentrate			Culture			Ratio: Per cent positive culture/Per cent positive concentrate	Concentrate			Culture			Ratio: Per cent positive culture/Per cent positive concentrate
	Total	Positive		Total	Positive		Total	Positive			Total	Positive		Total	Positive		
		Number	Per cent		Number	Per cent		Number	Per cent			Number	Per cent		Number	Per cent	
1940	606	367	60.5	1,893	326	17.2	520	208	40.0	2.3	586	26	4.4	586	160	27.3	6.2
1941	567	350	61.7	1,793	420	23.4	502	180	35.9	1.6	737*	43	5.8	735	221	30.1	5.2
1942	616	340	55.2	1,654	376	22.7	592	236	39.8	1.8	1,095*	85	7.8	1,079	268	24.8	3.2
1943	548	227	41.4	1,473	297	20.1	623	213	34.2	1.7	920*	38	4.1	912	215	23.6	5.8
Total.....	2,337	1,284	55.1	6,813	1,419	20.8	2,237	837	37.4	1.8	3,338	192	5.8	3,312	864	26.2	5.0

* A few gastric specimens strongly positive on concentrate were not cultured.

constant character of the patient group during the period of study. The slight decline in percentage over the four year period is probably a result of the gradual increase in the number of accidentally discovered cases, a trend also noticeable in table 1.

A more reliable index of the relative efficacy of the different methods of detecting tubercle bacilli can be obtained by determining which method was responsible for the first detection of acid-fast organisms in each patient admitted. The processes of selection inherent in the routine method employed again introduce a significant error since the first two specimens examined are usually sputum and are rarely cultured. A positive culture, therefore, frequently implies failure of at least three microscopic examinations.

TABLE 2

Efficiency of culture method: Results of culturing sputum and gastric specimens which were negative on concentrate

YEAR		TOTAL SPECIMENS CULTURED	POSITIVE ON CULTURE	
			Number	Per cent
1940	January to June	438	158	36.1
	July to December	554	165	29.8
1941	January to June	553	167	30.2
	July to December	541	171	30.8
1942	January to June	769	221	28.8
	July to December	704	191	27.1
1943	January to June	732	212	28.0
	July to December	673	184	27.4
Total		4,964	1,469	29.6

The results of this analysis are shown in table 3, which again reveals the importance of the cultural method. Of 611 patients with pulmonary tuberculosis admitted to this institution during a four year period, and whose sputum or gastric content was found to be positive for acid-fast bacilli at some time during the sanatorium stay, 65.8 per cent were first found to be positive by a microscopic method, while 34.2 per cent were first found by culture. This represents an increment by culture of about 50 per cent, since in virtually all these cases the microscopic methods had failed to detect the organism. Examination by cultural methods is thus responsible for the first positive finding in one-third of the cases found positive by all methods employed. Concentration methods account for an additional third, and direct smear the remainder. In this connection it

should be noted that examination of sputum by direct smear in this institution reveals acid-fast bacilli in only one-third of the patients with pulmonary tuberculosis in spite of the fact that great precautions are taken to choose a satisfactory sputum particle for smear.

The results presented in table 3 also serve to emphasize the importance of the examination of the gastric contents in the routine search for tubercle bacilli. It may be seen that 22.9 per cent, or almost one out of every four cases, is found to be positive for the first time by this method. Gastric examinations are frequently performed after repeated examination of the sputum, often with culture as well as concentration, has been unsuccessful, or when little or no sputum is available. Thus a large proportion of minimal and treated cases is included. By far the largest number of cases first found to be positive for acid-fast bacilli by examination of the gastric contents is discovered by culture and the overall increment by microscopic examination of the concentrate (5.2

TABLE 3

Analysis of type of examination which resulted in first detection of acid-fast bacilli

YEAR OF ADMISSION TO SANATORIUM	NUMBER OF PATIENTS FOUND TO BE POSITIVE BY SOME METHOD DURING SANATORIUM STAY	SPUTUM						GASTRIC			
		Smear		Concentrate		Culture		Concentrate		Culture	
		Number	Per cent	Number	Per cent	Number	Per cent	Number	Per cent	Number	Per cent
1940	160	57	35.7	45	28.1	29	18.1	8	5.0	21	13.1
1941	158	53	33.6	40	25.3	32	20.2	5	3.2	28	17.7
1942	158	49	31.0	48	30.4	17	10.8	13	8.2	31	19.6
1943	135	49	36.3	29	21.5	23	17.0	6	4.5	28	20.7
Total.....	611	208	34.0	162	26.6	101	16.5	32	5.2	108	17.7
				471	77.1					140	22.9

per cent) is so small as to be hardly worth the time spent in studying the slides. It has become a routine practice, therefore, to examine microscopically only those gastric specimens which can reasonably be expected to be positive on clinical and roentgenographic grounds, or on the basis of previous bacteriologic examinations.

All methods combined account for positive findings in only 86.2 per cent of the patients studied; in the remaining 13.8 per cent all specimens were negative on repeated examination (table 4). Unfortunately, guinea pig inoculations were not done on most of these cases but, wherever done, were also negative. This group of 98 cases is analyzed in table 5. About one-third (35 per cent) are patients in whom effective collapse therapy had been instituted prior to admission and whose sputum was presumably positive for tubercle bacilli before such therapy was begun. An additional third consists of patients with inactive disease, as evidenced by serial roentgenograms (28 per cent), and a few cases of pleural effusion without demonstrable parenchymal infiltration (5 per cent). A

few patients whose lesions were definitely proved to be nontuberculous and several patients with sarcoidosis of the lungs have not been included. Only a relatively small proportion of these negative cases (32 per cent), therefore, consists of patients with active disease, and of these the majority are of minimal extent and/or show very slight roentgenographic evidence of instability. If the other

TABLE 4

Incidence of cases persistently negative during sanatorium stay

YEAR	ADMISSIONS*	CASES FOUND POSITIVE BY SOME METHOD DURING SANATORIUM STAY		CASES PERSISTENTLY NEGATIVE DURING SANATORIUM STAY	
		Number	Per cent	Number	Per cent
1940	179	160	89.4	19	10.6
1941	179	158	88.3	21	11.7
1942	189	158	83.6	31	16.4
1943	162	135	83.3	27	16.7
Total	709	611	86.2	98	13.8

* Patients proved to be nontuberculous or those in whom the bacteriologic workup could not be considered adequate are excluded.

TABLE 5

Analysis of cases bacteriologically negative throughout sanatorium stay

YEAR OF ADMISSION TO SANATORIUM	CASES PERSISTENTLY NEGATIVE DURING SANATORIUM STAY*	ACTIVE DISEASE: LESIONS ROENTGENOLOGICALLY UNSTABLE DURING SANATORIUM STAY					EFFECTIVE COLLAPSE THERAPY INSTITUTED PRIOR TO ADMISSION				INACTIVE DISEASE: LESIONS ROENTGENOLOGICALLY STABLE DURING SANATORIUM STAY		PLEURAL EFFUSION WITHOUT DEMONSTRABLE PARENCHYMAL INFILTRATION	
		Total		Extent			Total		Type					
		Number	Per cent	Min.	Mod. Adv.	Far Adv.	Number	Per cent	Pneumothorax	Thoracoplasty	Number	Per cent	Number	Per cent
1940	19	4	21	4	0	0	11	58	10	1	4	21	0	—
1941	21	6	29	4	0	2	8	38	6	2	4	19	3	14
1942	31	8	26	6	2	0	8	26	6	2	14	45	1	3
1943	27	13	48	5	7	1	7	26	5	2	6	22	1	4
Total	98	31	32	19	9	3	34	35	27	7	28	28	5	5

* Cases proved to be nontuberculous are excluded.

groups are eliminated, these 31 patients represent only 4.8 per cent of the 642 patients with active pulmonary tuberculosis admitted to the sanatorium during the four year period. Acid-fast bacilli are thus demonstrable by microscopic or cultural methods in 95 per cent of patients in whom such a finding can reasonably be expected.

As it is reasonable to suppose that a certain proportion of the remaining 5 per

cent would have been found positive if a larger number of specimens had been examined, it is important from a practical standpoint to determine what procedures constitute a satisfactory bacteriologic investigation. The 611 positive cases were therefore analyzed to determine how many specimens had to be examined before a positive specimen was found by any method, and the results are tabulated in table 6. It will be noted that one-half (51.5 per cent) of the cases were found to be positive for acid-fast bacilli on the first examination. This usually included direct smear and concentrate, as well as a small proportion of gastric examinations (including culture) in those patients who either did not bring up sputum or had not yet learned how to do so. The high percentage of positive findings in the third specimen was largely a result of the fact that this specimen was cultured routinely if the preceding specimens were negative. Of particular significance is the fact that 82.5 per cent of the cases were found to be positive after three examinations (including at least one culture if the microscopic methods yielded negative results). Each subsequent examination added a very small increment. In all these respects the figures show a remarkable degree of constancy for the various yearly periods.

COMMENT

The importance of the culture method in examinations for acid-fast bacilli is clearly indicated by all the forms of analysis followed. In this institution the contribution of the use of culture was of a high order as it provided an increment of 30 per cent of positive findings in cases found negative for acid-fast bacilli by microscopic methods. Moreover, the use of cultures was the means of detecting the first acid-fast bacillus in one-third of the cases found positive by all methods combined. These particular values, however, are significant only within the routine framework employed and may not be extended to other situations without consideration of several variable factors.

The character of the patient group: The role played by culture becomes increasingly important as the number of bacilli in the specimen diminishes toward and beyond the limit of sensitivity of the microscopic method used. The present series includes a considerable number of patients with early and minimal lesions, as well as a significant number convalescing from surgical procedures such as thoracoplasty. Of this group only 66 per cent were first detected by the microscopic methods. In contrast to this, in a series of 507 patients analyzed by Pinner and Werner (1), over 60 per cent were classified as far advanced, and over 90 per cent were first found positive for acid-fast bacilli by the use of microscopic methods. Similarly, in 82 per cent of a series of 1,657 adult patients recently reported by Sanford (2), the first detection of acid-fast bacilli was made by microscopic methods. The death rate during the sanatorium treatment of this group was 34 per cent. Culture plays its greatest role in the identification of the relatively rare organisms in the pulmonary secretions of early, healing, and surgically treated cases, and therefore becomes progressively more important as the number of such cases increases in any patient group.

The quality of the microscopic methods: The performance of the culture methods

serves in some measure as a criterion of the quality of the smear and concentrate as well as of the efficiency of the culture. Since an increment by culture of 30 per cent (table 2) represents positive specimens which were not detected by the microscopic methods, it is reasonable to assume that an increase in the quality and efficiency of these methods should reduce the percentage. The various refinements aimed at accomplishing this purpose are directed either toward examination of a larger amount of the sample, or toward improvement of concentration and staining techniques. In institutional work, at least, all such refinements must be evaluated by weighing the additional time expended in the examination against the increase in positive findings that can be expected.

One of the methods of increasing the amount of the sample examined is the preparation and examination of more than one smear. Such a study was made in this laboratory using a series of 508 sputum concentrates. Duplicate smears were made and each was intensively examined for five minutes, thus doubling the usual examining time. The results, as presented in table 6, show an increment of 12 per cent in the percentage of specimens positive for acid-fast bacilli. Pottenger (3) has made an analysis of results obtained by examining sputum smears for increasing periods of time. Of sputum specimens microscopically positive after a fifteen minute search, 67 per cent were positive after five minutes and 90 per cent after ten minutes. This represents an increment of about 25 per cent for the second five minutes, or about twice that found in the duplicate smear series. It should be noted, however, that the criteria of positivity are not entirely comparable as culture is used in one series and microscopy in the other. In any event, the results of culture indicate that at least 45 per cent of the sputum concentrates examined are actually positive for tubercle bacilli, representing an increment about four to seven times as great as either method alone (table 6). Furthermore, in institutional practice at least, increasing the number of specimens examined tends to be more rewarding than increasing the time expended on each individual sample regardless of whether one or multiple smears are used.

Improvements in concentration technique have been limited by the necessity for retaining viable tubercle bacilli for subsequent culture. For this reason, most of the suggested improvements employing various hydrocarbon detergents cannot be readily utilized. Of the numerous modifications of staining technique that have been suggested, most of them offer little or no real advantage over the Ziehl-Neelsen method. A promising exception, however, appears to be the fluorescence method. In our own hands (4) little advantage was observed with the possible exception of the very thin gastric concentrate smears, but this appears to have resulted in large measure from an inefficient light source (5). More recent reports have been more enthusiastic (6).

The efficiency of the culture media: The number of suggested improvements in culture media are too numerous to detail here. Of the several tried in this laboratory, none so far suggested appears to offer much advantage over the modified Petragagni used. In spite of the disadvantages of malachite green as an inhibiting dye (7) it has, in our hands, offered the most satisfactory balance be-

TABLE 6

Analysis of number of specimens examined before first detection of acid-fast bacilli (all methods combined)

YEAR OF ADMISSION TO SANATORIUM	NUMBER OF PATIENTS FOUND TO BE POSITIVE AT SOME TIME DURING SANATORIUM STAY	FIRST ACID-FAST BACILLUS FOUND ON SPECIMEN NUMBER																	
		1		2		3		4		5		6		7		8		9 or more	
		Number	Per cent	Number	Per cent	Number	Per cent	Number	Per cent	Number	Per cent	Number	Per cent	Number	Per cent	Number	Per cent	Number	Per cent
1940	160	82	51.2	24	15.0	24	15.0	7	4.4	6	3.8	5	3.1	0	—	5	3.1	7	4.4
1941	158	84	53.1	15	9.5	32	20.2	8	5.1	6	3.8	5	3.2	5	3.2	0	—	3	1.9
1942	158	82	51.9	28	17.7	25	15.8	6	3.8	9	5.7	3	1.9	3	1.9	0	—	2	1.3
1943	135	67	49.6	20	14.8	21	15.5	10	7.4	5	3.7	4	3.0	2	1.5	2	1.5	4	3.0
Total...	611	315	51.5	87	14.2	102	16.7	31	5.1	26	4.3	17	2.8	10	1.6	7	1.2	16	2.6
		504	82.5																

tween growth of tubercle bacilli and the inhibition of contaminating organisms. Some index of the efficiency of the medium is provided by culture of specimens known to be positive for acid-fast bacilli on microscopic examination. Of 261 such specimens 221, or 84.7 per cent, showed growth. On the other hand, of all cultures planted, 5.1 per cent were discarded because of gross contamination.

There is obviously considerable room for improvement in the culture medium: in increasing the number of positive specimens; in reducing the number of contaminations; and in reducing the time required to produce visible growth. The synthetic medium containing water soluble esters of long chain fatty acids described by Dubos and others (8, 9, 10) appears to be a hopeful step in this direction. In its present form, however, it is not readily adaptable for routine use, especially in view of the frequency of contamination.

TABLE 7

Results obtained by doubling the examination time compared with those to be expected by culture

	TOTAL SPECIMENS EXAMINED	POSITIVE AFTER FIVE MINUTES		POSITIVE AFTER TEN MINUTES		INCREMENT BY DOUBLING TIME OF EXAMINATION	POSITIVE ON CULTURE		INCREMENT BY CULTURE OVER 5 MINUTE EXAMINATION
		Number	Per cent	Number	Per cent	*per cent*	Number	Per cent	*per cent*
Sputum concentrates examined by duplicate smear method	508	126*	25	141†	28	+12	239‡	47	+88
Sputum concentrates total for four year period	6,813	1,419	21	1,890§	28	+25	3,015‡	44	+110

* Figure represents number of positive findings on first smear examined.
† Figure represents number of positive findings on at least one of the two smears.
‡ Calculated, representing number of specimens positive after five minutes plus 30 per cent of the remaining negative specimens; the latter represents the average expected increment by culture as shown in table 2.
§ Calculated, representing the approximate number of positive findings to be expected after ten minutes based on the figures of Pottenger (3).

Time and frequency of use of culture methods: It is obvious that earlier and more frequent use of culture in any routine procedure tends to increase the number of positive findings and to reduce the average number of specimens required to demonstrate acid-fast bacilli. Thus, earlier use of culture would cause a "shift to the left" in table 5; its elimination would tend to produce a "shift to the right;" the examination of more specimens would be necessary before the same results are obtained No generalization as to the optimal time and frequency of use of culture can be made but, in view of the fact that such a large proportion of the first two specimens examined is found to be positive microscopically, earlier routine use of culture, in institutional work at least, is wasteful of time and culture media far out of proportion to any advantage gained. Modifications of the

routine can be readily made, however, if the laboratory is kept informed of existing problems, and close cooperation between laboratory and clinic may save a great deal of time and effort on both sides.

SUMMARY

An analysis of the results of routine examination for acid-fast bacilli of sputum and gastric contents over a four year period clearly demonstrates the important contribution of the cultural method within the routine framework employed. Culture is at least two to five times as effective as the microscopic methods in spite of selective weighting of the figures. In addition, it provides an increment of 30 per cent of positive findings in specimens found negative by the microscopic methods used and is a means of first detecting acid-fast bacilli in one-third of cases found positive by all methods combined. The importance of culture of gastric contents is emphasized, this method accounting for the first positive finding in 23 per cent of cases. All of these figures show remarkable constancy for each of the yearly periods. Of those cases found positive at some time during their hospital stay, 82 per cent were revealed by the first three examinations. Of all 709 patients studied, 14 per cent remained negative on repeated examination. Two-thirds of these patients, however, had either been previously treated or were roentgenologically inactive. If these cases are eliminated, acid-fast bacilli were demonstrated by microscopic and/or cultural methods in 95 per cent of patients in whom such a finding could be reasonably expected.

SUMARIO

Examen Sistemático del Esputo en Busca de Bacilos Acidorresistentes

Un análisis de los resultados obtenidos con el examen sistemático del esputo y el contenido gástrico en busca de bacilos acidorresistentes durante un cuatrienio demuestra claramente el importante aporte del método cultural en relacián con el sistema empleado. El cultivo resulta a lo menos dos a cinco veces más eficaz que las técnicas microscópicas a pesar de la valuación selectiva de las cifras. Además, suministra un incremento de 30 por ciento de hallazgos positivos en los ejemplares que resultan negativos con las técnicas microscópicas usadas y ofrece un medio de distinguir los primeros bacilos ácidorresistentes en la tercera parte de los casos que resultan positivos con todas las técnicas combinadas. Recálcase la importancia que revisten los cultivos del contenido gástrico, correspondiendo a esta técnica el primer hallazgo positivo en 23 por ciento de los casos. Todas estas cifras muestran notable constancia para cada uno de los períodos anuales. De los casos que resultaron positivos en alguna época durante su hospitalización, 82 por ciento fueron descubiertos durante los primeros tres exámenes. De los 709 enfermos estudiados, 14 por ciento permanecieron negativos a través de exámenes repetidos; pero dos terceras partes de éstos o bien habían sido tratados antes o eran radiográficamente negativos. Eliminando estos casos, se encontraron bacilos ácidorresistentes con las técnicas microscópicas y/o culturales en 95 por ciento de los enfermos en los que cabía razonablemente esperar dicho resultado.

Acknowledgment

The authors wish to thank Dr. Gabriele Bondi for her aid in analyzing the negative cases.

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A STUDY OF THE BACTERIOSTATIC AND BACTERICIDAL ACTIVITY OF PROMIN, DIASONE, SULFATHIAZOLE, SULFAGUANIDINE AND STREPTOMYCIN ON THE H-37V. STRAIN OF M. TUBERCULOSIS[1]

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INTRODUCTION

The trials that have been conducted in the chemotherapeutic treatment of tuberculosis in the past few years have revealed that only partial success is at hand. It is evident that it is not enough to obtain clinical cure through the use of antibiotic or other therapeutic drug even if the clinical improvement is dramatic. Because of the nature of the pathology of tuberculosis, it seems that any chemotherapeutic agent to be most effective in tuberculosis must have bacteriostatic as well as bactericidal properties. The exact action of the substances which have been used therapeutically in recent years is not clearly known and there has been insufficient work done on these substances in the laboratory, especially in relation to their bactericidal properties on *M. tuberculosis*. The present report concerns an investigation of the bacteriostatic and bactericidal properties of promin, diasone, sulfathiazole, sulfaguanidine, and streptomycin. The strain of *M. tuberculosis* of human origin H-37v. was the one chosen for the study.

In these studies the bacteriostatic effect was determined by the growth of the bacilli in a liquid culture medium to which various amounts of the drug to be tested were added to a series of culture tubes. The culture media used consisted of a modified Proskauer and Beck medium and the Dubos Tween-albumin medium. The growth of the bacilli was determined after twenty days of incubation at 37.5°C.

RESULTS

The bacteriostatic effect of the drugs tested is shown in table 1. It will be noted that streptomycin was the most, and sulfaquanidine the least, effective bacteriostatic agent of the drugs tested. In another experiment not shown in the table it was found that the addition of penicillin to the streptomycin dilutions did not materially enhance the tuberculostatic effect of streptomycin.

From these experiments it would seem that streptomycin and sulfathiazole gave the greatest promise for a possible favorable chemotherapeutic effect.

The bactericidal effect of the various compounds was determined by exposing a suspension of tubercle bacilli which contained one mg. (wet weight) of organisms per cc. of normal saline solution to different concentrations of the substance to be tested.

At one, twenty-four and forty-eight hour intervals after the bacilli were brought in contact with the drug, one cc. of the bacillary suspension was removed and washed in 20

[1] From the laboratory of J. C. R. S. Sanatorium, Spivak, Colorado.

volumes of normal saline solution by centrifugation for twenty minutes at 3,000 r.p.m. Cultures were made from the centrifuged sediment which was resuspended in the original volume. Guinea pigs were inoculated with 0.5 cc. of the washed bacillary suspension. Cultures and guinea pig inoculations were made from a control bacillary suspension which contained the same amount of tubercle bacilli but no drug. The cultures on egg-yolk-potato medium were examined at three weeks and the guinea pigs were killed on the day when the control animals died.

The results of these experiments are presented in table 2. As may be seen in the table, it was found that promin was distinctly bactericidal in solution of 100

TABLE 1

Bacteriostatic effect of various drugs on growth of tubercle bacilli in modified Proskauer and Beck broth (20 days incubation)

CONCENTRATION OF DRUGS	STREPTOMYCIN	SULFATHIAZOLE	PROMIN	DIASONE	SULFAGUANIDINE
mg. per 100 cc.					
100	—	XXXX	XXXX	XXXX	XXXX
50	—	XXXX	XXXX	XXXX	XXX
40	—	XXXX	XXXX	XXX	XXX
30	—	XXXX	XXX	XXX	XX
20	—	XXXX	XXX	XX	X
15	—	XXXX	XX	X	0
10	—	XXXX	X	0	0
5	—	XXXX	0	0	0
2.5	—	XXX	0	0	0
1	XXXX	X	0	0	0
0.1	XXXX	—	—	—	—
0.05	XXXX	—	—	—	—
0.02	XX	—	—	—	—
0.01	X	—	—	—	—
0.005	0	—	—	—	—
Control	0	0	0	0	0

Explanation of symbols (bacteriostatic effect):
XXXX = Complete bacteriostasis; no growth on culture medium.
XXX = Trace of growth.
XX = Poor growth.
X = Good growth.
0 = Heavy growth.
— = No test.

mg. per 100 cc. and diasone in 333 mg. per 100 cc., whereas the other compounds revealed no bactericidal effect. Streptomycin on the other hand reduced the number of colonies as determined by culture but did not kill all the organisms. In another experiment not shown in the table the addition of 100,000 units of penicillin per 100 cc. of the bacillary suspension did not alter the bactericidal effect of streptomycin.

The relative bacteriostatic and bactericidal properties as determined by *in vitro* studies and by animal inoculations on a streptomycin-sensitive strain of tubercle bacillus are presented in table 3.

TABLE 2

Bactericidal effect of various drugs on tubercle bacilli (cultures and animal inoculations were done with washed bacillary suspensions)

CONCENTRATION	DRUGS	CULTURE AFTER HOURS OF CONTACT WITH DRUGS			GUINEA PIGS INOCULATED WITH
		1 hr.	24 hrs.	48 hrs.	0.5 cc. of 48 hr. sample
mg. per 100 cc. 1,000	Streptomycin	—	xx	xx	x
500	Promin	0	xxxx	xxxx	xxxx
	Diasone	0	xxxx	xxxx	xxxx
400	Streptomycin	—	xx	xxx	x
333	Promin	0	xxxx	xxxx	xxxx
	Diasone	0	xxxx	xxxx	xxxx
250	Promin	0	xxxx	xxxx	xxxx
	Diasone	0	xx	xxx	xx
200	Streptomycin	—	xx	xx	x
100	Promin	0	xxx	xxxx	xxxx
	Diasone	0	x	xx	xx
	Sulfathiazole	0	0	0	0
	Sulfaguanidine	0	0	0	0
	Streptomycin	—	x	xx	x
Bacillary control suspension		0	0	0	0

Explanation of symbols (bactericidal effect):

xxxx = No growth on egg yolk medium: no tuberculous lesion in guinea pigs.

xxx = One or two colonies on culture: only local or lymph node lesion in guinea pigs.

xx = Few colonies on culture: moderate number of tuberculous lesions in spleen and liver.

x = Good growth on culture: generalized tuberculosis but not so extensive as in controls.

0 = Heavy growth on culture: massive generalized tuberculosis in guinea pigs.

— = No test.

TABLE 3

Comparative bacteriostatic and bactericidal efficiencies of various drugs on M. Tuberculosis

DRUGS	BACTERIOSTATIC CONCENTRATION IN MG. PER 100 CC.		BACTERICIDAL CONCENTRATION IN MG. PER 100 CC.
	Minimal inhibition	Complete inhibition	48 hours exposure to drugs
Promin	10	40	100
Diasone	15	50	333
Sulfathiazole	1	5	Absent
Sulfaguanidine	20	100	Absent
Streptomycin	0.01	0.05	?

(?)—Some bacilli were killed but some were viable after exposure to all concentrations; 10:1,000 mg. per 100 cc.

DISCUSSION

It is well known that the therapeutic efficacy of many drugs does not correspond necessarily with their bacteriostatic or bactericidal activity as found in *in vitro* studies. This is true in some of the sulfonamide compounds studied. For example, sulfathiazole manifested bacetriostatic activity *in vitro* which was eight times that of promin. Nevertheless, sulfathiazole was not bactericidal for *M. tuberculosis* when tested in concentrated solution for forty-eight hours and had no therapeutic effect on experimental tuberculosis in guinea pigs when 100 mg. were given daily. Diasone was found to possess less bacteriostatic and bactericidal potency than promin *in vitro*, but was almost as effective as promin in the treatment of tuberculous guinea pigs. According to Ballon and Gueron (1), the growth of *M. tuberculosis* was inhibited in a concentration of 100 mg. per 100 cc. of promin and in 10 mg. per 100 cc. of sulfathiazole. In the writer's laboratory the growth of *M. tuberculosis* H-37v. was completely inhibited in synthetic broth containing either 40 mg. per 100 cc. of promin or 5 mg. per 100 cc. of sulfathiazole. This difference may have resulted from the method of sterilization or from differences in the culture media employed. A few colonies of *M. tuberculosis* have been observed in a modified Herrold medium containing 50 mg. per 100 cc. of promin. The streptomycin exhibited the highest degree of bacteriostasis for it completely inhibited the growth of *M. tuberculosis* in a concentration of 0.05 mg. per 100 cc. Streptomycin appears to have little bactericidal effect, however, for it did not kill all the bacilli when tested in a concentration of 1,000 mg. per 100 cc. for forty-eight hours. The present observations agree with Middlebrook and Yegian (2) and Wolinsky and Steenken (3) relative to the bacteriostatic effect of streptomycin. The combination of penicillin and streptomycin in the proportion of one unit to one microgram, respectively, did not appear to have any influence on the effect of streptomycin. Since streptomycin is bacteriostatic in high dilution and is not bactericidal even in high concentration, it would seem reasonable to expect that a smaller dosage of the antibiotic could be used without sacrificing the therapeutic effect provided the tubercle bacilli remain sensitive. Fisher (4) found that cultures from one-half of patients treated with streptomycin showed various degrees of resistance to the antibiotic after 84 to 126 days of therapy. Cultures obtained from the other patients remained sensitive to streptomycin concentrations of one γ per cc. or less. In the J. C. R. S. Sanatorium streptomycin-resistant strains of tubercle bacilli have been cultured from over 70 per cent of the treated cases.

The problems that have arisen as the result of the therapeutic application of various chemotherapeutic agents in human tuberculosis suggest that the bactericidal, as well as the toxic and bacteriostatic properties of any new compounds, be determined by *in vitro* and *in vivo* laboratory studies prior to application to man. It may well be that a bacteriostatic agent without bactericidal properties may influence the appearance of resistant strains of organisms during a course of treatment.

SUMMARY

From laboratory studies with a streptomycin-sensitive strain of *M. tuberculosis* H-37v. it has been found that: (1) promin and diasone possess both bacterio-

static and bactericidal properties, with promin the more active of the two; (2) sulfathiazole possesses bacteriostatic activity to a high degree without appreciable bactericidal effect; (3) sulfaguanidine has much less bacteriostatic effect than sulfathiazole and is not bactericidal; (4) streptomycin exhibits the greatest bacteriostatic effect but has only slight bactericidal activity; (5) the bacteriostatic effect of streptomycin is not altered by the presence of penicillin.

SUMARIO

Estudio, con la Cepa H-37v del Bacilo Tuberculoso, de las Propiedades Bacteriostáticas y Bactericidas de la Promina, la Diasona, el Sulfatiazol, la Sulfaguanidina y la Estreptomicina

Por los estudios de laboratorio ejecutados con una cepa estreptomicinosensible del bacilo tuberculoso H-37v, se ha descubierto que: (1) la promina y la diasona poseen propiedades tanto bacteriostáticas como bactericidas, siendo la promina la más activa de las dos; (2) el sulfatiazol muestra alta actividad bacteriostática sin apreciable efecto bactericida; (3) la sulfaguanidina ejerce mucho menor efecto bacteriostático que el sulfatiazol y no es bactericida; (4) la estreptomicina manifiesta el mayor efecto bacteriostático pero apenas posee actividad bactericida, y (5) el efecto bacteriostático de la estreptomicina no es alterado por la presencia de penicilina.

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James Alexander Miller
1874–1945

James Alexander Miller
1874–1948

Dr. James Alexander Miller, a former president of the National Tuberculosis Association and the New York Tuberculosis and Health Association, died on the 29th of July, 1948, at the age of seventy-four.

After completing his preliminary education in Princeton University and receiving his medical degree from the College of Physicians and Surgeons of Columbia University in 1899, he entered upon the practice of medicine in New York City, continuing until the time of his death. Starting in 1901, he also practiced for eight summers at Paul Smith's in the Adirondacks, and it was here that he cultivated a long lasting friendship with Dr. Edward Livingston Trudeau and acquired the inspiration to concentrate much of his later professional activity in the field of tuberculosis. During the subsequent years he extended his interest into other sides of medicine and public health, but always with particular focus on the unique problems of this specific disease. His growing experience and the time tested attributes of his personality, intellect, and character qualified him as an able leader and marked him for succession to many posts of responsibility and importance. His merits included a love of the truth, high ideals, devotion to study, patience, perseverence, self-discipline, generous consideration of his associates, and easy adaptability.

Doctor Miller's interest and accomplishments in the campaign against tuberculosis were not defined by any narrow limits, since he was a man of versatility and naturally endowed with broad vision. Early in his career he became interested in the Tuberculosis Service, now the Chest Service, of Bellevue Hospital, which at the time hardly deserved the name of a clinical service. Through the ensuing years he succeeded in creating a competent organization which raised the standard of medical work to its present level. Recognizing the numerous backgrounds of tuberculosis, he interested philanthropic laymen in the social problem, leading to the organization of an Auxiliary which raised funds from private sources and looked after many of the needs of the impoverished patients. He became Professor of Clinical Medicine in the College of Physicians and Surgeons of Columbia University and developed a course of instruction for medical students and for the house staff of the hospital which continues to claim the interest of many young physicians at the start of their careers. In the 1920's Doctor Miller obtained funds to initiate research as a feature of the Service; this too has grown and continues to the present date. This connection with Bellevue brought him into contact with many governmental officials who respected his judgment and advice, and it was largely through his personal efforts and influence that the present building housing the Bellevue Chest Service was planned and constructed.

The public health aspects of tuberculosis also commanded his interest and energy. Aside from his work in the national and local tuberculosis associations, he was an officer or adviser in many organizations, including the Community

Service Society, the Milbank Memorial Fund, the Commonwealth Fund, and the New York Academy of Medicine where, for many years, he was chairman of the Committee on Public Health, now Public Health Relations. His counsel was often followed because of his unusual ability to visualize problems in concrete terms, to plan the logical and practical approach, and to set in motion the machinery for solution. In action he never spared himself and it was largely for this reason that he was so successful in obtaining the cooperation of others. In his large private practice it was his humanity which impressed so many patients.

The wide recognition of Doctor Miller's abilities and accomplishments was indicated in part by his election to the presidency of such medical organizations as the American College of Physicians, the American Climatological and Clinical Association, the Practitioner's Society, the New York Medical and Surgical Society, and the New York Academy of Medicine. From 1927 to 1945 he also served as President of the Board of Trustees of the Trudeau Sanatorium. He continued as Visiting Physician in Charge of the Tuberculosis Service of Bellevue Hospital until 1938, following which he was Honorary Consultant. He was honored with Doctor of Science degrees by Columbia (1930) and Princeton (1936) and with a Doctor of Public Health degree by New York University (1937). In recognition of his work with the Red Cross and the Rockefeller Commission for the control of tuberculosis in France during World War I, he was made a Chevalier of the Legion of Honor. The National Tuberculosis Association awarded him the Trudeau Medal in 1944 and the New York Academy of Medicine, its Academy Medal in 1947. On this occasion, marking its Centennial Celebration, the honor was bestowed upon him as "the Academy's most distinguished and beloved Fellow and one of the greatest benefactors of mankind."

J. Burns Amberson

THE

AMERICAN REVIEW OF TUBERCULOSIS ABSTRACTS

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Bronchial Stenosis after Pneumothorax.—The authors report 3 cases of pulmonary tuberculosis in which stenosis of the left main bronchus developed about two and a half years after the beginning of pneumothorax treatment. These constrictions within the bronchus were smooth, no ulceration being present. Biopsies showed they were composed of fibrous tissue without characteristic microscopic signs of tuberculosis. Clinically the patients had at first improved under pneumothorax treatment but eventually had suffered from recurrent febrile attacks following retention of sputum and dilatation of the bronchus below the constriction. In one case multiple areas of softening developed throughout the collapsed lung. In another a subapical cavity increased in size. In the third the constriction was not followed by changes in the collapsed lung. It was possible in each case to locate the constriction by tomography and confirm its presence by bronchoscopy. The use of lipiodol was considered inadvisable. The stenoses were believed to be due to a sclerosing type of bronchial tuberculosis aided by mechanical factors. One of these may have been the narrowing of the lumen of the left bronchus by congestion of its network of arterial, venous and lymphatic vessels following pneumothorax. Such congestion could result ultimately in fibrosis. This form of stenosis can be considered a clinical entity which can be diagnosed by the presence of a smooth constriction of the left main bronchial stem, developing a year or two after the induction of pneumothorax and followed by febrile episodes with alternate retention and outpouring of sputum. Tomography is very useful in detecting and locating the constriction. As regards treatment, discontinuance of the pneumothorax is not always indicated since a lung blocked by stenosis will not reexpand and lack of air in the pneumothorax area may cause serious symptoms. Thoracoplasty is usually contraindicated. Dilatation of the stenosis may succeed but the only logical treatment is complete removal of the involved lung.—*Réflexions sur les sténoses des bronches souches sous pneumothorax artificiel, G. Giraud, A. Balmes, L. Nichet & J. Mirouze, Le Poumon, September-October, 1947, 3: 329.*—(*A. T. Laird*)

Bronchial Foreign Bodies.—In dealing with translucent foreign bodies diagnostic roentgenograms should be overpenetrated and the correct position determined by fluoroscopic examination. Both inspiratory and expiratory films should be made to show mediastinal shift. Proper roentgenograms may show a distinct interruption of the air column of the involved bronchus.—*Some observations on the roentgen diagnosis of nonopaque foreign bodies aspirated into the bronchi, S. Welin, Acta radiol., 1948, 29: 529.*—(*J. E. Farber*)

Metallic Breath Sounds.—Metallic breath sounds are usually heard over a pneumothorax space when inspiratory and expiratory pressures are both positive. Occasionally they can also be heard over a pneumothorax with negative pressures. A certain tension of the pneumothorax wall is necessary for the production of these metallic breath sounds and this tension can be due either to a positive pressure pneumothorax or to changes in the

structure of the chest wall itself due to inflammation, fibrosis, or shrinkage. Metallic sounds over a closed pneumothorax are faint and loud metallic sounds suggest the presence of a valvular pneumothorax or of a very large cavity in the lung. Metallic sounds may develop rapidly over a negative pressure pneumothorax when a dry pleurisy causes increased tension of the pneumothorax wall.—*Metallic über dem Pneumothoraxraum-kein obligates Ueberdrucksymptom, K. Bauer, Wien, klin. Wchnschr., December 12, 1947, 59: 811.*—(*G. C. Leiner*)

Accidental Extrapleural Pneumothorax.—Accidental extrapleural pneumothorax may occur during the course of therapeutic intrapleural pneumothorax as described in 1933 by Corbetta. Oeschsli in 1938 reported 15 instances among 400 pneumothorax cases. The author describes 7 cases seen within two years. The roentgenologic appearance is striking with gas in the extrapleural space. This gas is very slowly absorbed and may cause pain in the hypochondrium with a tight feeling in the chest. The extrapleural cavities may be confused with adhesions so that the pneumothorax is sometimes stopped. Manometric readings may be confusing and in themselves lead to abandoment of the pneumothorax. Finally, the extrapleural pocket may enhance the formation of adhesions by approximating the pleural surfaces.—*Accidental extrapleural pneumothorax, O. Olsson, Acta radiol., 1948, 28: 117.*—(*J. E. Farber*)

Level of Diaphragm in Emphysema.—The level of the diaphragm is of clinical importance in the diagnosis of emphysema. Radiological aids for this diagnosis are (*1*) position of the ribs, (*2*) translucency of the lung fields, and (*3*) the level of the diaphragm. None of these has been determined accurately and quantitatively. The level of the diaphragm was measured in this work. The distance between a line joining the iliac crest and the level of the highest point of the diaphragmatic dome on each side on full inspiration and full expiration was measured, as suggested by Warner and Doidge. This can be determined more accurately by screening at 3 ft. distance than by films, since under the fluoroscope one can check that the subject has actually reached the limit of inspiration or expiration. The quotients:

$$\frac{\text{distance from lowest diaphragmatic level to iliac crest}}{\text{distance from tuber ischii to acromion process}}$$

and

$$\frac{\text{distance from highest diaphragmatic level to iliac crest}}{\text{distance from tuber ischii to acromion process}}$$

were established. Altogether 58 subjects were examined, including 8 who were normal, 5 obese normal, 7 with heart disease who had recently suffered from congestive failure but were not in failure at the time, and 38 with clinical emphysema. These results show what was to be expected, namely, that the level of the diaphragm in pronounced emphysema is always lower than normal. In slight emphysema a tendency toward a low diaphragm is unmistakable although the level may be within normal limits. The only exceptions are the obese emphysematous patients, in whom the diaphragm would have to overcome the higher resistance of the more voluminous abdomen. It is possible that this resistance prevents the lowering of the diaphragm. Any tendency to increase the volume of the lung would, in these cases, result in the lifting of the ribs only. This is known to happen in many cases of emphysema apart from the diaphragmatic changes. The following mean values were secured using the aforementioned quotients (first figure is "low level" of diaphragm, second figure is "high level"): normals, 0.35, 0.47; slight emphysema, 0.31, 0.44; moderate and severe emphysema, 0.27, 0.34; obese emphysema, 0.37, 0.47; obese normals, 0.36, 0.48; cardiac patients recovered from congestive failure, 0.36, 0.44.—*Radiological determination of the level of the diaphragm in emphysema, M. Grossmann & H. Herxheimer, Brit. J. Radiol., September, 1948, 21: 446.*—(*B. Hyde*)

Loeffler's Syndrome with Cavity Formation.—A 39 year old woman developed several pulmonary infiltrations within a few weeks, with fever and a blood eosinophilia of 30 per cent. The infiltrations resorbed promptly but one of them showed cavity formation in the centre. This cavity healed within three weeks.—*Ueber ein zerfallendes, eosinophiles, flüchtiges Lungeninfiltrat, A. Ott, Wien. klin. Wchnschr., May 21, 1948, 60: 320.*—(*G. C. Leiner*)

Pleural Effusion.—The object of this study was to estimate the significance of early pleural effusion in pulmonary embolism. In a series of 46 cases, 21 were found to have a pleural effusion. In more than one-half of these cases the pleural effusion was more evident roentgenographically than the parenchymal density. However, the presence of the effusion was of little help in diagnosing pulmonary embolism. In a series of 38 cases of pneumonia and bronchopneumonia only a few developed pleural effusion and the parenchymal density was the principal radiographic finding. Pleural effusions are sometimes seen in acute, non-specific respiratory tract infections without parenchymal lesions or with only small lung densities.—*Early pleural effusion in pulmonary embolism and pneumonia or bronchopneumonia, G. Moberg, Acta radiol., 1948, 29: 7.*—(*J. E. Farber*)

Spastic Bronchitis.—Bronchitis spastica without real asthmatic attacks has been seen more frequently recently than in previous years, possibly as a consequence of the war. The dyspnea which is found in spastic bronchitis can always be relieved by epinephrine. Bronchospasm is always seen in patients with emphysema and is frequent in cardiac patients.—*Ueber Bronchospasmus, W. Gröger, Wien. klin. Wchnschr., January 23, 1948, 60: 49.*—(*G. C. Leiner*)

Mediastinal Hernia.—Mediastinal hernias may be caused by pressure-difference in the two hemithoraces at the 2 weak places described by Nitsch. Hernias caused by pulsion (pneumothorax) are demonstrated by fluoroscopy. Hernias caused by traction are best shown by tomography. The diagnostic signs seen in the tomograms are the upper borderline of the hernia and the visualization of the blood vessels crossing from the healthy hemithorax into the pouch of the hernia. (Author's Summary)—*The roentgen examination of the mediastinal lung hernia with reference to tomography, H. Salinger, Acta radiol., 1948, 29: 130.*—(*J. E. Farber*)

Thoracic Aortography.—The author reports a new technique for visualization of the thoracic aorta. A catheter is introduced into the radial artery and guided under radioscopic control via the subclavian artery into the ascendent aorta. After the rapid injection of 30 cc. of diodrast, exposures of the aorta are made. (Author's Summary)—*Thoracal aortography by catheterization from the radial artery, S. Radner, Acta radiol., 1948, 29: 178.*—(*J. E. Farber*)

Thoracic Aortography.—A method for aortography suggested by Radner and somewhat modified has been used in 3 cases. The authors consider that the method should be preferred to angiocardiography at contrast roentgen examinations of the aorta and its intra-thoracic branches. (Authors' Summary)—*Thoracic aortography, B. Brodén, H. E. Hanson & J. Karnell, Acta radiol., 1948, 29: 181.*—(*J. E. Farber*)

Lung Changes in Dermatitis.—In 2 patients with an acute generalized dermatitis roentgenograms of the lung showed an increased reticular pattern. As the skin lesion cleared, the abnormal lung changes disappeared. The author suspects hypersensitivity as the cause of both lesions with changes probably in the interstitial tissue.—*On lung changes in acute general dermatitis, N. Frostberg, Acta radiol., 1948, 29: 493.*—(*J. E. Farber*)

Mediastinal Abscess.—Roentgenologic diagnosis of a mediastinal abscess can some-

times be aided by mediastinal puncture and injection of a contrast medium. The risk of such a puncture is reduced by using a very fine needle and the procedure is much easier if performed under fluoroscopy. In the case reported, the patient was given a little barium to swallow to simplify the localization. The needle was inserted into the right supraclavicular fossa in a downwards and medial direction reaching the abscess wall. The cavity was aspirated and penicillin was injected with subsequent improvement.—*Mediastinal abscess diagnosed by roentgen, S. R. Kjellberg, Acta radiol., 1948, 30: 294.—(J. E. Farber)*

Tracheal Diverticula.—Multiple tracheal diverticula may be demonstrated by bronchography, as occurred in one patient being studied for bronchiectasis. Besides the diverticula, cylindric and saccular bronchiectasis was found in both lower lobes.—*A case of multiple tracheal diverticula, K. Nielsen, Acta radiol., 1948, 30: 331.—(J. E. Farber)*

BCG and Tuberculosis Diagnosis.—Following a BCG vaccination the tuberculin test gives a positive reaction and, therefore, cannot be used further as an aid to differential diagnosis. A Russian method of avoiding this difficulty was tested. In a group of 180 vaccinated children the Mantoux reaction was positive in 86 per cent; the Pirquet test, if performed simultaneously, was positive in 52.8 per cent. In another group of 180 vaccinated children the Pirquet test was done first and the Mantoux test was done two weeks later. There was a positive Mantoux reaction in 86 per cent of the children but a positive Pirquet reaction in only 5.8 per cent. None of the 360 children had pathological changes in the sedimentation test or the roentgenologic examination. It is believed that the Mantoux and Pirquet tests, if done at a certain interval, will be of use in the differential diagnosis. A positive Pirquet reaction after BCG vaccination does not mean the presence of a reinfection nor is reinfection excluded by a negative Mantoux and a negative Pirquet reaction.—*Experimentelle Beiträge zu den in Zusammenhang mit der B. C. G.—Impfung auftauchenden differentialdiagnostischen Problemen, St. Zimányi, Wien. klin. Wchnschr., August 13, 1948, 60: 515.—(G. C. Leiner)*

Auto-urine Injections for Tuberculosis.—Eight patients with pulmonary tuberculosis received intramuscular injections of their own fresh urine. The treatment was started with 0.5 cc., the injections being given every two or three days. The dose was increased every time by 0.5 cc. to a final volume of 5 cc. No improvement of the pulmonary condition of the patients was seen.—*Ergebnisse der Eigenharnbehandlung nach Plesch bei der Lungentuberkulose, Beyrer, Wien. klin. Wchnschr., July 2, 1948, 60: 418.—(G. C. Leiner)*

Ultrasonic Waves and Tubercle Bacilli.—Human tubercle bacilli can be damaged by exposure to ultrasonic waves so that they stop multiplying. These tubercle bacilli no longer cause tuberculosis in the guinea pig but, if injected, they produce some immunity to subsequent infection with virulent tubercle bacilli. It can be expected that tubercle bacilli which have been exposed to ultrasonic waves will be used for prophylactic vaccination of human beings. *Ueber die Wirkung von Ultraschall auf Tuberkulosekeime von Typus humanus and die Möglichkeit einer Schutzimpfung gegen die Tuberkulose, F. Kress, Wien. klin. Wchnschr., September 17, 1948, 60: 597.—(G. C. Leiner)*

Pulmonary Edema.—The roentgenologic picture of pulmonary edema, acute and chronic, has only recently been described. The heart is enlarged and the lung fields contain symmetrical, confluent, perihilar ill-defined patches of increased density. The central distribution of the opacities leaving the apices, peripheral borders, and bases relatively clear is characteristic. The bronchi stand out clearly whereas the vascular markings are not seen. The differential diagnosis includes

bronchopneumonia or multiple hemorrhagic infiltration of pleura and lungs. In the three cases of verified pulmonary edema presented the roentgenographic changes appeared earlier than the clinical signs.—*On the roentgenologic picture of pulmonary edema, S. Rennocs, Acta radiol., 1948, 30: 169.*—(*J. E. Farber*)

Tuberculoproteins. I.—Different preparations of purified protein derivative and Old Tuberculin are known to differ in their skin activity. Since this property is associated only with protein, a partial explanation of this phenomenon is that the protein content of certain lots of Old Tuberculin may be significantly different but this can hardly account for the variation found in preparations of PPD. The presence of more than one protein or the degradation of the protein during preparation might account for the observed facts. Concentrated, unheated culture filtrates of two strains of human tubercle bacilli, a virulent and a slightly virulent one, were fractionated with ammonium sulphate to give 14 fractions from each strain. Chemical determinations and sedimentation velocity measurements were made on those fractions for which significant results could be obtained. The evidence showed that two distinct proteins are present in addition to a polysaccharide and nucleic acid. The physical measurements did not demonstrate the presence of any other proteins. One of the proteins was isolated in pure form and found to have a molecular weight of 44,000 ± 5,000, as determined by the partial specific volume, sedimentation velocity, and diffusion rate. This protein is believed to be the one previously isolated by Seibert et al., who assigned it a molecular weight of 32,000. The other protein was not freed from polysaccharide so that its molecular weight could not be determined although it is believed to have a sedimentation constant of about 2 S. Sedimentation and diffusion constants were obtained for the polysaccharide which appears to be a homogeneous molecular species with a molecular weight of about 20,000.—*The proteins in unheated culture filtrates of human tubercle bacilli I. Fractionation and determination of physical-chemical properties, E. B. Bevilacqua & J. R. McCarter, J. Exper. Med., March, 1948, 87: 229.*—(*J. S. Woolley*)

Tuberculous Droplet Infection in Rabbits.—Experiments have been reported in which rabbits were made to inhale virulent bovine tubercle bacilli, air-borne as separated cells in fine drop nuclei under standardized conditions. The tubercles which developed in their lungs were known to be induced by organisms derived from single cells, all of which were implanted upon alveolar tissue within relatively brief intervals. For the first five or six weeks the rate of tubercle development was found to be uniform and not appreciably affected by the number of the lesions, their position in the lungs, or by the varying susceptibility of the rabbits. This initial, homogeneous phase contrasted sharply with the later stages of the infection which were strikingly heterogeneous especially when rabbits of various strains were used. These features of air-borne tuberculosis are now reported in more detail. The Ravenal strain of *Mycobacterium tuberculosis bovis* was prepared in an aqueous suspension which was whirled into a fine mist in a special aerosol flask. Only the smallest droplets evaporated rapidly enough to produce the droplet nuclei of which the final aerosol consisted. Occasional droplet nuclei contained tubercle bacilli. The concentration of the organisms in the aerosol and the length of exposure determined the intensity of infection. Fifty-six albino rabbits were exposed in these experiments and killed at various intervals. During the first week after infection the bacilli were always found in isolated alveolar macrophages. Between the sixth and ninth days the number of infected macrophages rapidly increased and at twelve days there were collections of parasitized macrophages occupying one or two alveoli. At this time leucocytes and small monocytes had begun to accumulate about the developing tubercles. A brief, early nonspecific response to uninfected droplet nuclei also was seen. By the

end of the third week typical tubercles became visible. During the fifth week differences in the rate of progress of the tubercles from animal to animal were detectable although all lesions were remarkably uniform in size in the lungs of any one animal. By the end of the sixth week of infection individual differences in the rate of progress of the initial tubercles and of the infection as a whole became evident. No evidence of delayed tubercle formation was encountered. This heterogeneity of the pattern of the disease seemed to be related to the intensity of the inflammation which in turn seemed to correspond to the differences in the growth rate of the bacilli after the fourth week, these differences being estimated by the number of organisms found in the lesions. It appears that heterogeneity of progressive tuberculosis may be related to differences in the capacity of animals to change the composition of their tissue or blood in such a way that growth of the bacilli may be more or less inhibited. It is concluded that rabbits do not differ in their lack of resistance to initial growth of bovine tubercle bacilli. However, the later, heterogeneous pattern of response suggests that these animals vary widely in their capacity to acquire resistance.—*Tuberculosis of rabbits induced by droplet nuclei infection: I. Initial response to infection, H. L. Ratcliffe & W. F. Wells, J. Exper. Med., June, 1948, 87: 575.*—(*J. S. Woolley*)

Apical Lung Cancer.—Experience with 92 cases of the Pancoast-Tobias tumor is presented. Early diagnosis can be made by roentgenograms and the first sign to be observed is a horn-like or semilunar density covering the pleural dome. Tomography helps to separate atelectasis from the tumor shadow. Several tumors varying both histologically and in location were found to produce the syndrome.—*Sur la pathologie et le diagnostic radiologique du cancer apical pulmonaire et des tumeurs malignes péri-apicales, R. Mathey-Cornat & de Fleurian, Acta radiol., 1948, 29: 19.*—(*J. E. Farber*)

Traumatic Paralysis of Diaphragm.—A 51 year old man fell from a ladder, fractured and dislocated his right clavicle, and fractured the upper seven ribs on the right. The right diaphragm was paralyzed, apparently by compression of the phrenic nerve between the fractured clavicle and the lateral processes of the cervical spine.—*Traumatische Zwerchfellähmung mit Totalverrenkung des Schlüsselbeins, R. Hagen, Wien. klin. Wchnschr., September 24, 1948, 60: 624.*—(*G. C. Leiner*)

Hering-Breuer Reflex.—The minimal pressure required for a reflex respiratory response during pentothal inhibition may be used to measure the rate of recovery of excitability. Reflex respiratory responses to light pressure on the chest wall in the deeply anesthetized dog are abolished by bilateral cervical vagotomy. Unilateral cervical vagotomy or unilateral pneumonectomy enhances the reflex response of the deeply anesthetized dog when lying on the unoperated side; when the operated side is down the reflex response is depressed. The dog with a unilateral vagotomy or unilateral pneumonectomy has a faster respiratory rate when lying on the unoperated side than when on the operated side. This difference in rate with change in position persists indefinitely in the unanesthetized animal. (Authors' summary).—*Role of the Hering-Breuer reflex under deep pentothal anesthesia, J. C. Scott, E. A. Reed, D. Saris & H. P. Redondo Ramirez, Am. J. Physiol., September, 1948, 154: 428.*—(*G. C. Leiner*)

Rib Anomaly.—The chest roentgenogram of a 25 year old female showed an unusual distal bifurcation of the fourth rib on the right. This bifurcation caused a constriction of the right hemithorax, a pleural adhesion, and the development of a septum running through the middle and lower lobes of the lung. This is the so-called "bifid rib of Luschka".—*Ueber eine ungewöhnliche rippenanomalie, zugleich ein Beitrag zur distalen Rippengabelung, P. Lutz, Wien. Kl in. Wchnschr., December 7, 1947, 59: 846.*—(*G. C. Leiner*)

Tracheal Foreign Body.—The author describes a previously unreported sign of foreign

bodies in the trachea in children. There is inspiratory collapse below and simultaneous widening of the air passages above the foreign body. These changes disappear on expiration.—*A previously unreported sign of foreign bodies in the trachea, S. R. Kjellberg, Acta radiol., 1948, 30: 800.*—(*J. E. Farber*)

Thymic Shadow on Chest Roentgenogram.—This report is based on 498 consecutive roentgenograms of children first taken when they were six months old and subsequently at six month intervals. A peculiar sail-like, triangular projection was noted extending out from the mediastinum in 44 children (8.8 per cent). In 12 patients the shadow was so large that it obscured almost the whole of the upper lung field. There were 12 children with large shadows, 12 with medium sized shadows, and 20 with small shadows. In 31 cases the shadow was on the right side, in 12 cases it was on the left, and in one there was a small bilateral projection of which the right was the larger. In some cases the mediastinum was displaced towards the side of the shadow. Lateral roentgenograms were not of value but oblique films indicated that the shadow was anterior. In many cases, the lower edge of the shadow on the right side coincided with the plane of the horizontal fissure. Eleven of these children were followed for two years or more, during which time the shadows gradually diminished in size and in some cases disappeared. Almost all these children were in good health when roentgenographed, only 4 being ill. None had any symptom or sign of pulmonary or cardiac disease and postmortem correlation indicated that this shadow was actually thymus gland. In an infant, who died following a splenectomy for an obscure form of hemolytic anemia, the chest roentgenogram taken at autopsy revealed the previously described shadow on the right side. After the sternum had been removed but before the organs were disturbed, surgical clips were placed on the poles of the thymic lobes and a repeat chest roentgenogram was taken. It was found that the clips corresponded exactly with the outlines of the shadow seen on the first roentgenogram. In stillborn children, the lateral limits of the thymus extend farther out than when the lungs are inflated. It was suggested that, as the lungs expand, the thymus is stripped from the chest wall and compressed into the mediastinum, but that sometimes one or both lower poles are adherent so that it remains anchored in the position occupied in the fetus. This theory was supported by the observation that in some cases with a unilateral shadow the mediastinum shifted toward that side. As the child grows older, the thymus is gradually stripped back until the shadow ultimately disappears. The finding of this sail-like, triangular projection from the mediastinum (thymus) does not have any pathologic significance.—*A sail-like triangular projection from the mediastinum: A radiographic appearance of the thymus gland, F. H. Kemp, H. N. C. Morley & E. Enrys-Roberts, Brit. J. Radiol., December, 1948, 21: 618.*—(*L. Hyde*)

Roentgenograms in Sarcoid.—The roentgenological aspect of sarcoidosis is discussed. Lung films may show enlargement of mediastinal nodes, miliary infiltration, areas of fibrosis, and confluent areas of patchy infiltration. Bilateral mediastinal lymphadenopathy is common but unilateral involvement occurs. Similarly, unilateral parenchymal lesions occur but also less often than bilatetal lesions. Pulmonary manifestations are rarely uniform; no single type of infiltration can be described as an early or late phase of the disease. Reisner, however, believes that the disseminated nodular forms are early lesions. Skeletal changes occur in 20 per cent of the patients. The bone lesions resemble small cysts and are found in the phalanges of the fingers and toes.—*Roentgenological aspect of sarcoidosis, A. J. Ackerman, Am. J. Roentgenol., March, 1948, 59: 318.*—(*J. E. Farber*)

Retrocardiac Bronchiectasis.—Bronchiectasis involves the lower lobe of the left lung more frequently and more extensively than any other pulmonary area. The occurrence of unilateral bonchiectasis is nearly five times as

frequent in the left as in the right lung. Isolated bronchiectasis of the retrocardiac area of the left lower lobe is frequent. On routine chest roentgenograms it may escape recognition entirely and can be diagnosed only by bronchographic examination.—*Retrocardiac bronchiectasis, R. G. Bloch, L. F. Sandock & E. B. Mitchell, Am. J. Roentgenol., August, 1948, 60: 219.*—(*J. E. Farber*)

Streptomycin in Tuberculosis Surgery.—Prior to the use of streptomycin, pulmonary resections for tuberculosis carried a mortality rate of 25 per cent and an incidence of empyema of 12 per cent and of bronchopleural fistula of 8 per cent. In most places, the incidence of spread following thoracoplasty was 3 to 6 per cent. In 28 cooperating hospitals every other tuberculous patient undergoing thoracoplasty and every patient with tuberculosis undergoing resection received streptomycin. The daily dosage at first was 2.0 Gm.; later this was reduced to 1.0 Gm. The drug was given for one week before and two weeks after each operation. In a total of 1,347 thoracoplasty stages, streptomycin was given for 699 operations on 258 patients. Postoperative spreads or reactivations appeared in 14 (2 per cent) and wound infections in 8 patients. In the control group, there was spread in 5.6 per cent and a correspondingly larger number of wound infections. The reduction was felt to be significant, but other factors such as improved technique, anesthesia, et cetera, may have contributed. Therefore, the routine administration of streptomycin in thoracoplasty is not recommended; it should be given in selected cases. A total of 129 resections were done, including 77 lobectomies and 52 pneumonectomies. There was a mortality rate of 4.5 per cent and an incidence of empyema of 4 per cent. These results are so striking when compared to the previous figures as to indicate that the use of streptomycin in pulmonary resection is mandatory.—*Streptomycin in surgery of pulmonary tuberculosis, J. D. Murphy, Surg., Gynec. & Obst., November, 1948, 87: 546.*—(*A. G. Cohen*)

Streptomycin for Tuberculosis.—Streptomycin was used in the treatment of 225 tuberculous patients after August, 1947, at the Hôpital Laennec in Paris. The average daily dose was 1.5 Gm. Skin eruptions appeared in 22 per cent, nausea and vomiting in 34 per cent, and vestibular disturbances in 39 per cent. There was one case of deafness which appeared after three months of treatment. Renal function could not be adequately studied. Therapeutic results were as follows: Of 78 patients with meningitis, 31 were living after six to eight months. Among 54 cases of miliary tuberculosis with or without meningitis, there were 36 surviving patients, mostly those without meningitis. Roentgenograms showed complete clearing. Of 64 patients with pulmonary tuberculosis (nonmiliary), 70 per cent had regression of clinical symptoms (fever, weight loss, cough, expectoration). Regressive roentgenographic changes were most marked in the diffuse nodular infiltrations of recent date; old lesions showed no response. Pneumonic consolidations sometimes gave a good initial response but the effect was usually incomplete. Cavitary lesions were seldom influenced but good results were obtained by combining streptomycin with collapse therapy. Thirty-three cases of laryngeal and pharyngeal tuberculosis showed favorable results with healing of the tuberculous lesions in 12 cases and great improvement in 11 cases. Dysphagia disappeared with great rapidity; the effect on dysphonia was slower and less constant. Streptomycin broke up the parallelism usually existing between the laryngeal and pulmonary lesions. Regression of laryngeal pathology was observed with no change in the pulmonary status. In endobronchial tuberculosis, streptomycin had a favorable influence on edematous, granulomatous and freshly ulcerative forms.—*Rapport sur le traitement de la tuberculose par la streptomycine, E. Bernard & A. Lotte, with the collaboration of B. Kreis, J. S. Bourdin & G. Arnaud, Rev. de la tuberc., 1948, 12: 165.*—(*V. Leites*)

INCIPIENT PULMONARY TUBERCULOSIS IN PERSONS OVER FORTY[1]

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INTRODUCTION

There is a rather widespread clinical impression that pulmonary tuberculosis rarely makes its appearance after the age of thirty. For example, in the most recent edition of an outstanding textbook of medicine (1) the following statement is found in the section on tuberculosis: "Those who have lived to the fourth decade without the appearance of a plumonary lesion seldom develop the disease." Nevertheless, it is the impression of many phthisiologists (2, 3, 4) that the first roentgenographic evidence of significant pulmonary tuberculosis after thirty, and even after forty, is not at all a rarity today. For instance, of the 344 cases of incipient pulmonary tuberculosis recently reported by Reisner (2), 17 developed after the age of forty. It was thought worthwhile, therefore, to report an additional 5 cases of incipient pulmonary tuberculosis in persons over forty years recently observed by the author.

Source of Material and Selection of Cases

The 5 cases reported here were observed by the author since his assignment in April, 1946 to the Kips Bay Health Center Chest Clinic, one of the twenty-one chest clinics of the Bureau of Tuberculosis, Health Department, New York City. This clinic averages approximately 2,000 adult "contact" examinations yearly. The established policy is to follow all adult contacts to active cases of pulmonary tuberculosis every six months with complete chest examination, including a roentgenogram, as long as contact is maintained and for two years after contact has been broken. With only few exceptions, the population served by the clinic is white and the overwhelming majority of the contacts supervised (the exact per cent is difficult to determine) are below forty years of age.

All 5 patients included in this study, which ended September, 1948, had recent "normal" chest films as "contacts" which were carefully reviewed by the author and all developed the first roentgenographic evidence of tuberculosis after their fortieth birthday. Except for age, the same criteria for inclusion in this group of cases were used as is described by Reisner in his articles (2).

CASE REPORTS

Case 1: L. P. (figures 1A and 1B) is a white male born in 1899. He was a contact to his wife who had apparently arrested far advanced pulmonary tuberculosis at the time of his first examination at the Kips Bay Clinic on April 16, 1942. This film was entirely "normal" (figure 1A). On the next clinic visit, February 21, 1945 (at which time patient was 46 years

[1] From the Bureau of Tuberculosis, New York City Department of Health, 125 Worth Street, New York 13, New York, and the Departments of Medicine and Preventive Medicine of the New York Hospital-Cornell University Medical College.

TABLE 1

Observations at the Time of Initial Demonstration of the Pulmonary Lesion
(5 cases personally observed combined with 17 cases from Doctor Reisner's series to make a total of 22)

MONTHS FROM LAST NORMAL CHEST ROENTGENOGRAM TO INITIAL LESION	TOTAL NUMBER	STAGE OF DISEASE AT FIRST APPEARANCE[1]			SYMPTOMS	
		Minimal	Moderately Advanced	Far Advanced	Present	Absent
4 to 6	9	7	2		3	6
7 to 12	10	6	2	2	3	7
13 to 24	1		1		1	
Over 24	2		1	1	2	
Total.....	22	13	6	3	9	13

[1] According to criteria of National Tuberculosis Association.

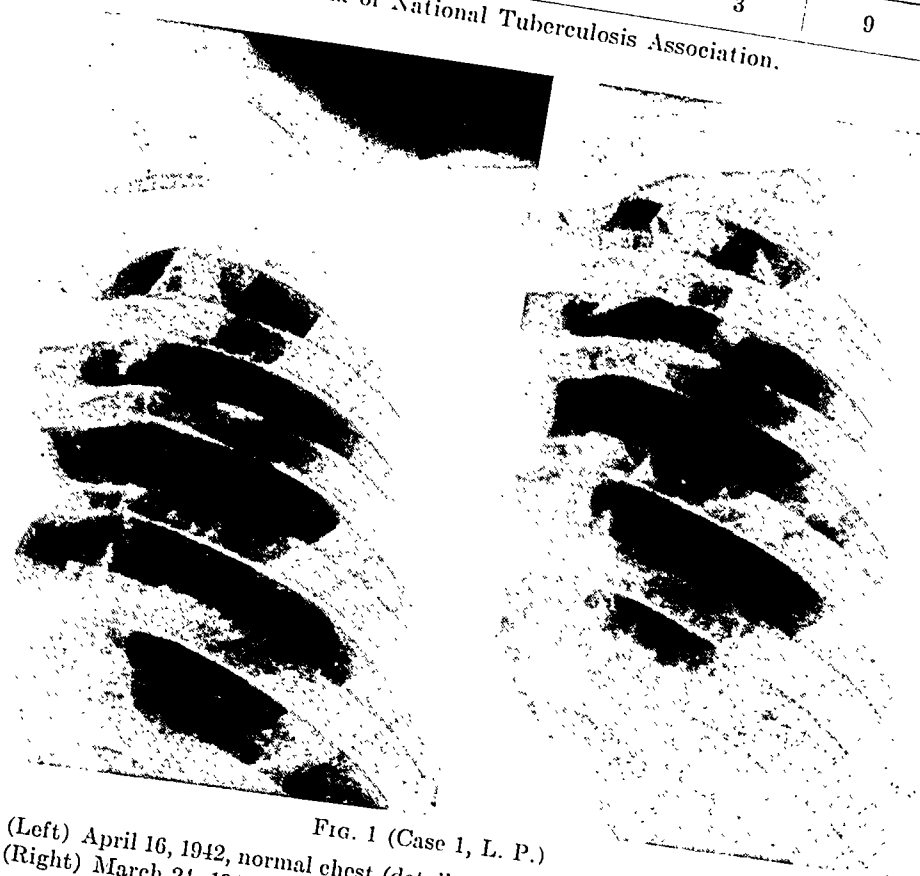

FIG. 1 (Case 1, L. P.)

A. (Left) April 16, 1942, normal chest (detail of left upper lung field).
B. (Right) March 24, 1948, area of infiltration in first a.i.c.s. on left. This lesion had been unstable between February 21 and May 29, 1945. No interval films available. Now considered arrested.

old) he complained of cough with the expectoration of blood-streaked sputum during the preceding two weeks. A chest roentgenogram, obtained at that time revealed definite infiltration in the left upper lung and the diagnosis of active, moderately advanced pulmo-

nary tuberculosis was made. The patient never placed himself under adequate treatment and very few sputum examinations were done (all negative for *M. tuberculosis*). Subsequent chest films on March 9, 1945 and May 29, 1945 revealed progression of infiltrations and probable cavitation in the left upper lung. The patient was lost from supervision until March

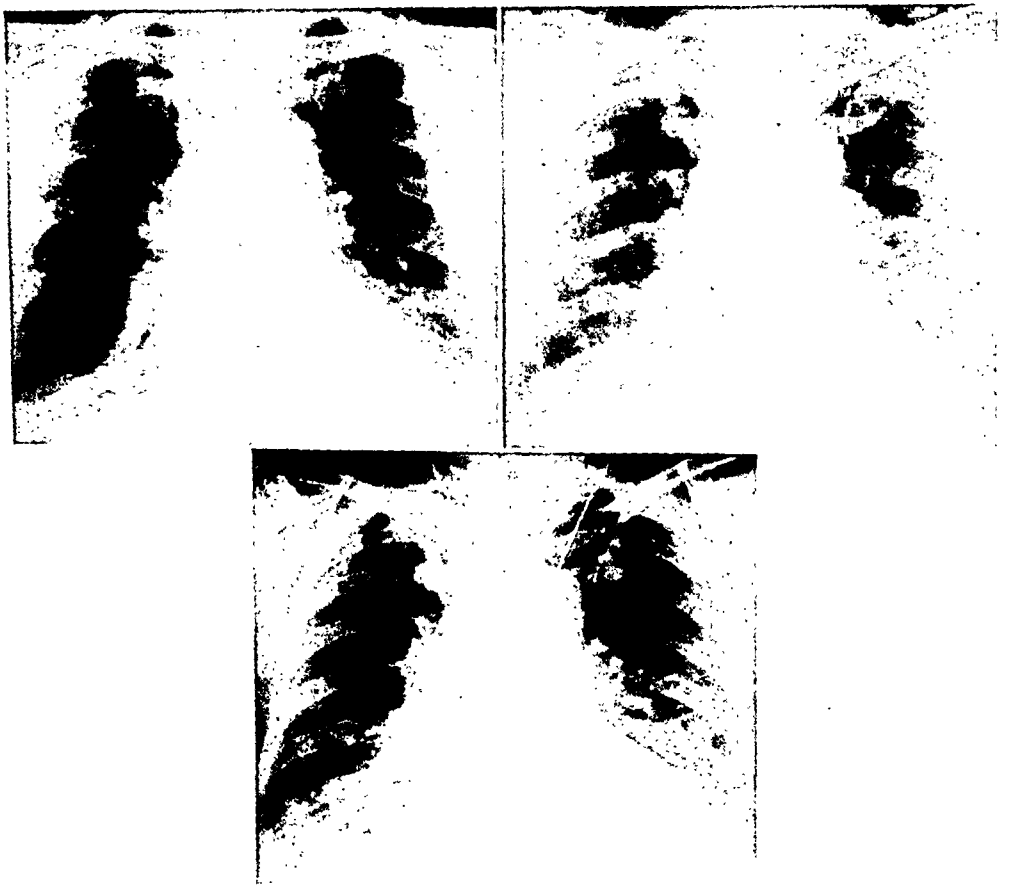

FIG. 2 (Case 2, M. W.)

A. (Upper left) January 23, 1946, cardiac enlargement and wide aorta (hypertensive) Lung fields considered normal. Calcification at right hilum.

B. (Upper right) January 31, 1947, round area of infiltration, periphery of first a.i.c.s. on right. In previous film of July 29, 1946, there was a larger area of infiltration which decreased considerably in size in the six month interval.

C. (Bottom) March 15, 1948, extension and breakdown of lesion in right upper lung field, between first and third anterior ribs. There is probably a small lesion below first rib on the left.

24, 1948, when a chest film (figure 1B) revealed clearing and "hardening" of the lesions, and he stated that he was entirely asymptomatic. The diagnosis was changed to arrested moderately advanced pulmonary tuberculosis. The total period of observation from first recognition of disease was three years.

Case 2: M. W. (figures 2A to 2C) is a white female born in 1886, with known hypertensive cardiovascular disease and mild diabetes, whose daughter died of pulmonary tuberculosis in August, 1947. The first chest roentgenogram at Kips Bay Clinic was taken on January

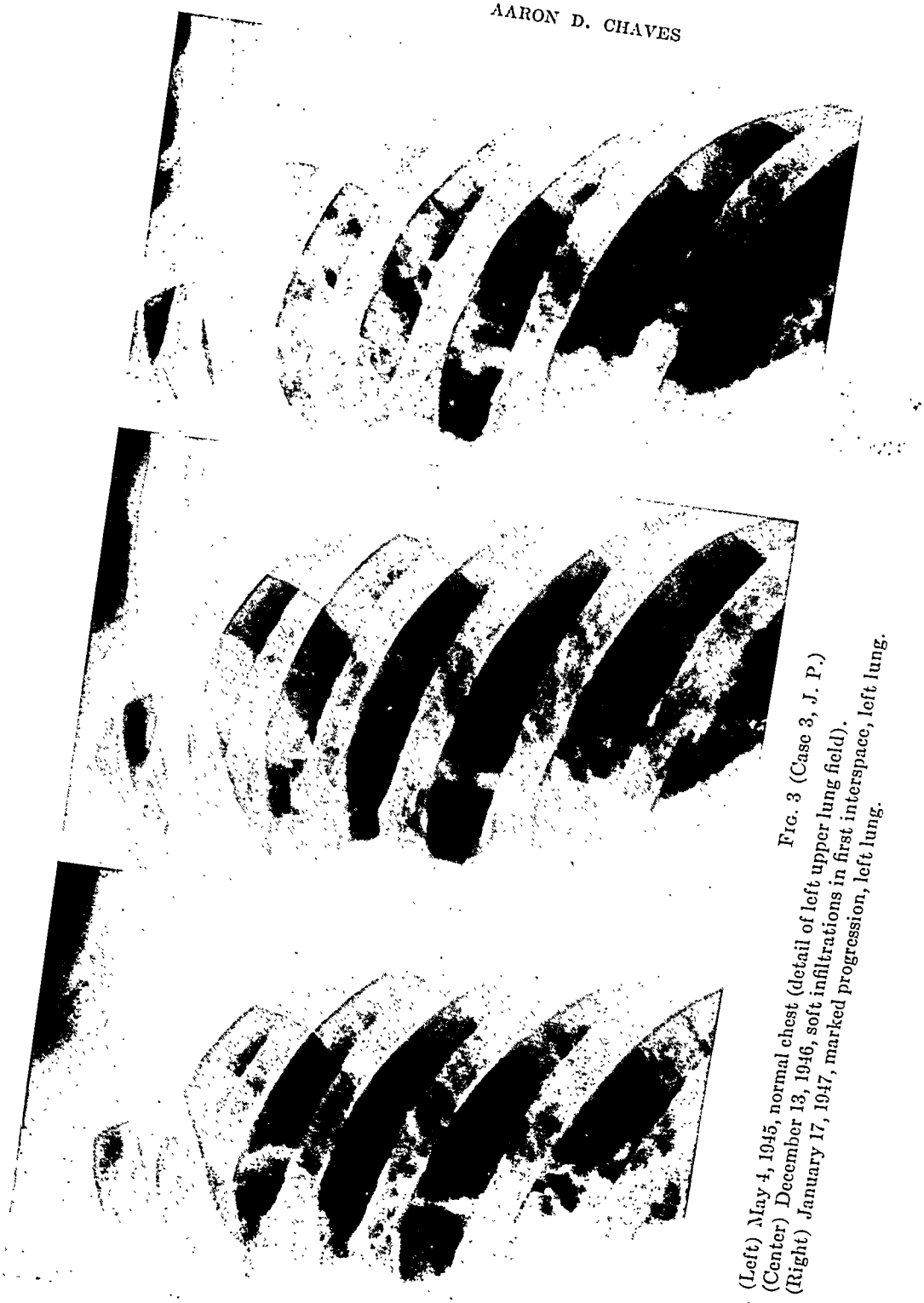

FIG. 3 (Case 3, J. P.)

A. (Left) May 4, 1945, normal chest (detail of left upper lung field).
B. (Center) December 13, 1946, soft infiltrations in first interspace, left lung.
C. (Right) January 17, 1947, marked progression, left lung.

23, 1946 and revealed an enlarged heart, a widened aorta and only a calcified primary complex in the lung (figure 2A). The next routine film of July 29, 1946 (when patient was 60 years old) revealed a round homogeneous density, 2 cm. in diameter, in the first intercostal space on the right. The density subsequently first decreased in size (figure 2B) and then showed definite progression (figure 2C). Sputum examinations from November, 1946 until September, 1948 were consistently positive for acid-fast bacilli on direct examination. The patient was admitted to Triboro Hospital in September, 1948 with moderately advanced cavitary disease. She always denied having symptoms and was extremely reluctant to accept hospitalization. The total period of observation was approximately two years, during which time the pulmonary tuberculosis progressed from a minimal to a moderately advanced state.

Case 3: J. P. (figures 3A to 3C) was a white male born in 1903 who had been heavily exposed to a friend who died of pulmonary tuberculosis in 1945. Roentgenograms obtained at the clinic between May 4, 1945 (figure 3A) and May 22, 1946 were all interpreted as normal. Routine films on November 27, 1946 and December 13, 1946 (when patient was 43 years old) revealed a very small soft infiltration in the first anterior intercostal space on the left (figure 3B). Patient was immediately recalled for sanatorium care, but did not report to the clinic until January 17, 1947, at which time his chest film revealed marked progression of the disease involving the upper half of the left lung field (figure 3C). He was promptly admitted to the New York Hospital, where his sputum was found to be positive for *M. tuberculosis*. He was given a course of streptomycin and transferred to a sanatorium after considerable improvement and disappearance of *M. tuberculosis* from the sputum. He soon had a reactivation of the disease in his left upper lobe, however, and the sputum again contained *M. tuberculosis*. The reactivated lesions did not respond to bed rest and a second course of streptomycin. An attempt at left pneumothorax was unsuccessful, so a first stage thoracoplasty was done in April, 1948. He died shortly after the operation. Postmortem examination revealed that an acute myocardial infarction was the cause of death. The total period of observation from discovery of minimal lesion to death was approximately sixteen months.

Case 4: S. D. (figures 4A to 4D) is a white female, born in 1890, who had had mild diabetes for many years and contact with an open case of pulmonary tuberculosis (husband) since March, 1942. She was examined regularly at the clinic, with normal chest roentgenograms between October 18, 1943 and May 20, 1946 (figure 4A). Films of May 20, 1946 and December 9, 1946 (when patient was 56 years old) revealed a very smal soft infiltration which was overlooked as it was covered by the posterior part of the fifth rib on right. A routine film of the chest on January 12, 1948 and re-examination on January 21, 1948 revealed a homogeneous exudative infiltration in the right upper lung (figure 4B). The patient was asymptomatic and refused hospitalization. Subsequent films in the clinic revealed marked progression of the disease with formation of a large cavity in right upper lung (figures 4C and 4D). Sputum concentrations and cultures were reported negative for *M. tuberculosis* until April 19, 1948. Since then, all sputum examinations have been positive for *M. tuberculosis*. The patient still denies symptoms and refuses hospitalization. Her diabetes is fairly well controlled by a regimen supervised by a diabetic clinic. The total period of observation from first appearance of the minimal lesion to the present far advanced bilateral pulmonary tuberculosis is nineteen months.

Case 5: T. M. (figures 5A and 5B) a white male born in 1895, was admitted to the clinic on August 22, 1947 as a contact to a friend who died in August, 1947 of tuberculosis. The first roentgenogram of the chest on August 22, 1947 was interpreted as normal (figure 5A). A routine film of February 20, 1948 (when patient was 53 years old) revealed a soft infiltration in the third anterior interspace on the right which had already showed considerable progression by March 3, 1948 (figure 5B). The patient was promptly hospitalized at Bellevue Hospital, where *M. tuberculosis* was found in the sputum. He was subsequently transferred to the Municipal Sanatorium at Otisville, where he has remained up to the present (September, 1948). The sputum is still positive for *M. tuberculosis*. The total period of observa-

tion from the initial demonstration of a minimal lesion to the present status, as active moderately advanced tuberculosis with tubercle bacilli in the sputum, is five months.

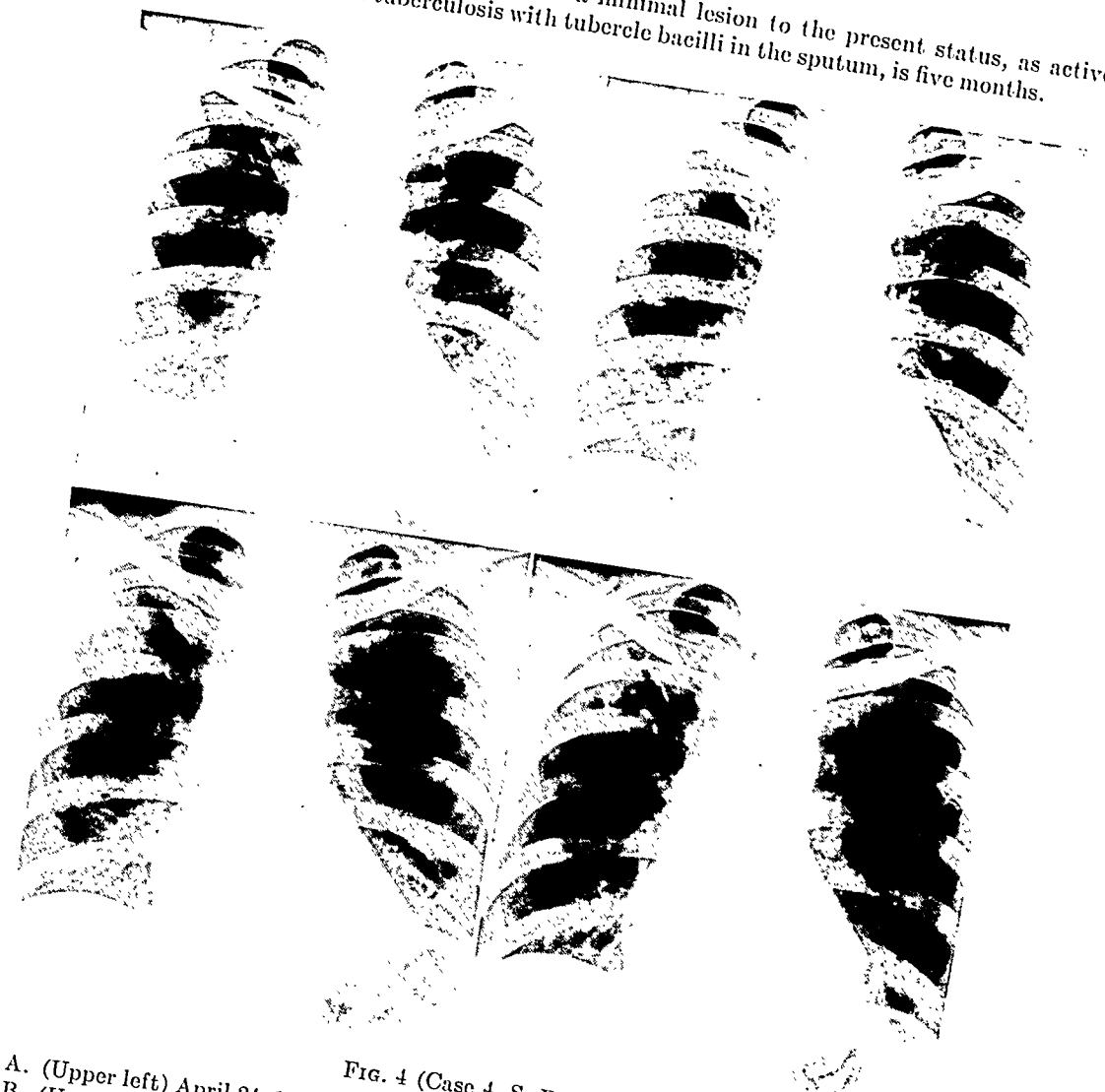

FIG. 4 (Case 4, S. D.)

A. (Upper left) April 24, 1944, normal chest.

B. (Upper right) January 21, 1948, soft infiltration, periphery of right lung, between first and second a.i.c.s.

C. (Lower left) April 19, 1948, marked progression on right. New lesion on left (behind intersection of third and sixth ribs).

D. (Lower right) July 14, 1948, further progression bilaterally, with cavity formation on right.

Analysis of Combined Series

It was thought worth while to combine these 5 cases with the 17 cases of pulmonary tuberculosis developing after forty years of age included in Reisner's large series of incipient pulmonary tuberculosis. This makes a group of 22 cases ob-

served in various New York City Health Department Clinics, for the most part, since 1940. One cannot expect to learn too much from analyzing such a small group of cases derived from material with the many shortcomings already discussed in Reisner's recent articles (2). Nevertheless there are in this group notable clinical features.

The ages varied from 40 to 75; 9 were male, 15 female; 17 were white and 5 nonwhite. All of the patients were known to have had recent intimate contact with active cases of tuberculosis. Five patients presented roentgenographic

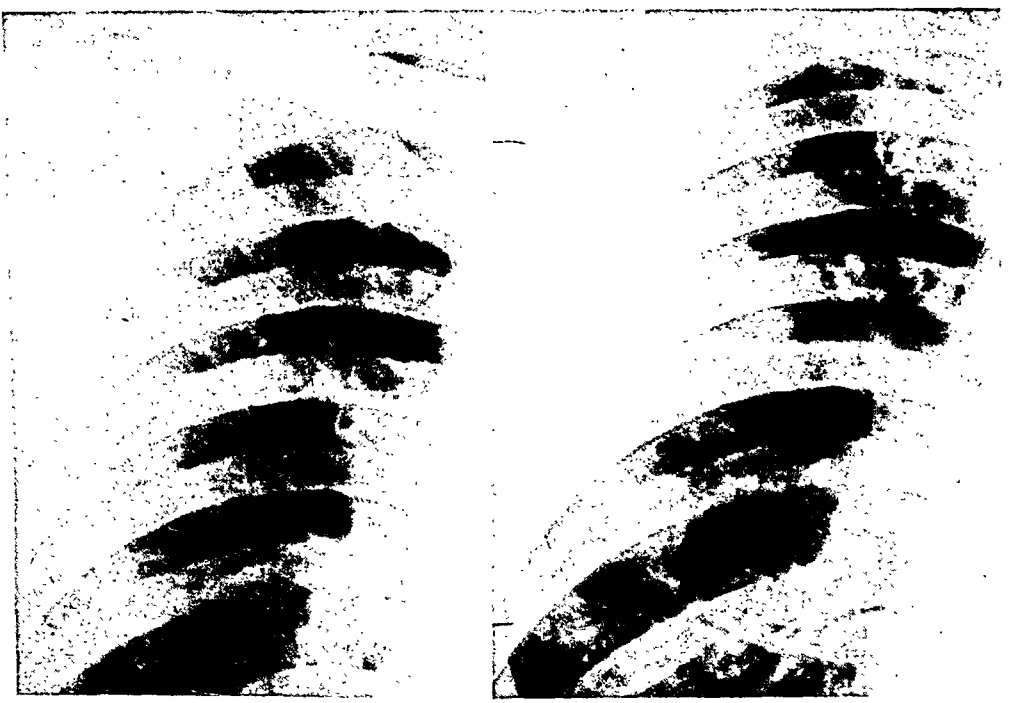

FIG. 5. (Case 5, T. M.)

A. (Left) August 22, 1947, normal film (detail of right upper lung field).
B. (Right) March 3, 1948, soft patchy infiltration in periphery of right lung between third and fourth a.i.c.s.

evidence of a healed, calcified, primary complex. No tuberculin testing was done on any of the patients prior to development of recognizable tuberculosis. In all but 5 instances, bacteriologic confirmation of the diagnosis was obtained at some time during the period of observation. The sputum investigations made early in the disease in most instances were not sufficiently intensive to be of much significance.

Observations at the time of initial demonstration of the pulmonary lesion: In table 1 are listed some of the observations made at the time of initial appearance of the pulmonary lesion. Nineteen cases were observed to develop disease within one year of a normal roentgenogram, while 3 (all with symptoms) were first discovered with disease approximately two, three and four and one-half years

respectively after a normal film. The latter were already advanced cases on discovery. While it is obvious that the earlier the lesion is discovered, the greater the chance of its being minimal, asymptomatic, and noninfectious, it is interesting to find that 2 nonwhite patients with moderately advanced disease with sputum positive for *M. tuberculosis* were encountered only six months after a normal film. One 45 year old white woman developed far advanced disease (fatal within one and one-half months after diagnosis) seven months after a normal film. Four of the 13 minimal cases were found to have *M. tuberculosis* in the sputum at the time of discovery.

As is to be expected, there were many cases in this group associated with other diseases. Four patients had mild, uncontrolled diabetes at the time of discovery, one of the diabetics also having hypertension and one arteriosclerotic heart disease. In his paper on early pulmonary infiltration, Amberson (5) mentions diabetes as one of the factors which predispose to the late development of progressive

TABLE 2

Observations at Conclusion of Period of Observation (September, 1948)

STAGE OF DISEASE AT INITIAL DEMONSTRATION	STATUS AT CONCLUSION OF OBSERVATION			
	Arrested	Active		Dead
		Minimal	Moderately or far advanced	
Minimal (13 cases)	4	2	4	3
Moderately advanced (6 cases)	4*			2
Far advanced (3 cases)				3
Total	8	2	4	8

* 2 cases with disease controlled with pneumothorax.

tuberculosis. However, none of the patients, as far as could be ascertained, had "dietary deficiencies, alcoholism and the like", also listed by Amberson as factors usually associated with late development of pulmonary tuberculosis. Three patients had hypertension with cardiac enlargement (one diabetic) and 2 had arteriosclerotic heart disease (one diabetic). One patient died of a myocardial infarction following a first stage thoracoplasty (J. P., Case 3). One patient had latent syphilis and one presented temporary manifestations of a psychosis.

Observations at conclusion of period of observation: The status of the 22 cases at the conclusion of the period of observation, which varied from six months to ten years and averaged 39 months, will be found in table 2. The striking finding in this small series is the extreme seriousness of the disease. Eight of the 22 patients have already died, and 4 patients are considered at present to have progressive and advanced tuberculosis with a rather poor prognosis. Only 8 cases are classified as arrested, 2 with the aid of pneumothorax. In the 8 fatal cases, duration from initial discovery to death varied from one and one-half months in a case which was far advanced to six and one-half years in a case which was mini-

mal at time of discovery. Two of the patients with minimal lesions died sixteen and seventeen months, respectively, after initial demonstration of the lesion, one from myocardial infarction and the other from rapidly progressive tuberculosis complicated by diabetes. Of the 4 diabetics, the tuberculosis of one is now well arrested, one is dead, and 2 have progressive and advanced tuberculosis with a poor prognosis. The type of treatment administered to the 22 patients varied considerably from prompt, excellent care, to no treatment at all.

COMMENT

One outstanding fact is evident from the study: a roentgenographically normal chest in a person over 40 does not eliminate the possibility of pulmonary tuberculosis developing in the future. As a matter of fact, incipient pulmonary tuberculosis in persons over 40 may be much more common than is generally supposed. From discussions with many observers, it is the author's impression that more and more cases of early tuberculosis in the older age group are being seen in various chest clinics throughout the country. Reisner (2) and Robins (4) have both called attention to the late appearance of the initial roentgenographic evidence of pulmonary tuberculosis in many cases under their observation, while both Pinner (3) and Reisner (2) have recently indicated the need for serial roentgenographic observations in the older age groups among the apparently healthy population to determine the frequency of incipient clinical tuberculosis in such groups.

It is impossible, of course, to draw any conclusions as to the pathogenesis of incipient clinical pulmonary tuberculosis in older people from this very small series of cases drawn from a highly selective sample of the population. Theoretically, late incipient pulmonary tuberculosis may come about as the result of: (*1*) reactivation of old roentgenographically undetectable foci (endogenous exacerbation); (*2*) new exogenous infection in individuals with a clinically healed primary complex as determined by a positive tuberculin test; (*3*) new exogenous infection in individuals with an obsolete primary infection which can be determined clinically by the reconversion of a previously known positive tuberculin test to negative (second "primary infection"); (*4*) recently acquired first infection (true primary infection).

The entire problem of pathogenesis might be clarified to some extent by determining the frequency with which incipient tuberculosis occurs in older individuals who are negative tuberculin reactors shortly prior to the initial demonstration of pulmonary tuberculosis. Unfortunately, there are no up to date data on the incidence of negative tuberculin reactors in the older age groups, and the need for such a study becomes obvious. Both Pinner (3, 6) and Rich (7) have indicated the need for large periodic tuberculin surveys of apparently healthy populations of all ages, *including those over 40 years of age.*

The importance of clear understanding of the pathogenetic mechanisms of pulmonary tuberculosis in the older age groups can be readily understood in view of the marked change in the epidemiological picture of the disease, with a striking shift of the mortality peak towards the older age groups. While

in 1900 only about 25 per cent of all tuberculosis deaths occurred after the age of 45 (3), at present 50 per cent of all tuberculosis deaths in this country occur after the age of 45 (8). If one considers the white males alone, over 75 per cent of tuberculosis deaths today (in New York City) occur after the age of 40 (9). Proper public health methods for the control of tuberculosis may well depend to a considerable extent on a better understanding of all the factors contributing to this remarkable shift in the peak mortality.

On the basis of the findings of this small and highly selective series, one gets the impression that, in the material available from a "contact" population, the incipient tuberculous lesions of persons over 40 show about the same tendency to progression as those under 40. The applicability of these observations to incipient lesions found among the general population remains to be determined.

SUMMARY

1. Five cases of incipient pulmonary tuberculosis in persons over 40 years of age are reported. All 5 patients had a recent normal chest roentgenogram.

2. Of this group of 5 cases, one died of a myocardial infarction following a first stage thoracoplasty, 2 have active progressive disease complicated by diabetes, one has active, moderately advanced tuberculosis, and the infection in one is arrested.

3. By adding these 5 cases to 17 cases of incipient pulmonary tuberculosis in persons over 40 observed by Reisner, a total of 22 cases are made available for study. Of this group, 8 are dead, 4 have active and progressive disease with a poor prognosis, 2 have active but minimal lesions, and the disease in 8 was considered to have been arrested at the conclusion of the period of observation.

4. The need for serial roentgenographic observations and repeated tuberculin surveys in the older age groups is discussed.

SUMARIO

Tuberculosis Pulmonar Incipiente en los Cuarentones

1. Comunícanse cinco casos de tuberculosis pulmonar incipiente en personas de más de cuarenta años. Los 5 enfermos habían mostrado recientemente una radiografía normal.

2. De los 5, uno ha muerto de infarto miocardíaco consecutivo al primer tiempo de una toracoplastia, 2 tienen tuberculosis evolutiva activa complicada con diabetes, uno tiene tuberculosis moderadamente avanzada, activa, y en uno la infección está estacionada.

3. Agregando estos 5 casos a los 17 de tuberculosis pulmonar incipiente observados por Reisner en personas de más de 40 años, cuéntase para estudio con un total de 22 casos. De este grupo, 8 han muerto, 4 tienen tuberculosis evolutiva y activa con mal pronóstico, 2 tienen lesiones activas, pero mínimas, y 8 parecían hallarse estacionados al terminar el período de observación.

4. Discútese la necesidad de ejecutar observaciones radiográficas seriadas y repetidas encuestas con tuberculina en los grupos de mayor edad.

Addendum

Since submitting this paper for publication four months ago, the writer has observed 9 additional cases of incipient pulmonary tuberculosis in persons over 40 years of age which fulfill the criteria for inclusion in this study. Two of the cases were seen at the Kips Bay Chest Clinic; 3 cases were observed on the Chest Service of the New York Hospital; one was seen on the Pulmonary Service of Montefiore Hospital; 3 cases were reviewed (charts and roentgenograms) from other New York City Health Department Chest Clinics.

Five patients were male, 4 were female; 6 were white and 3 were Negro. The interval between the last normal chest film and the initial roentgenographic evidence of disease varied from six months to ten years, with most intervals between six and twelve months. Ages of the patients at the time of the last normal chest films ranged from 40 to 62 years.

Six patients had definite symptoms antedating the discovery of disease, 2 of whom had significant exposure recently to tubercle bacilli. All 6 of these patients had far advanced active pulmonary tuberculosis with sputum positive for *M. tuberculosis* at the time of discovery. Three cases were discovered as the result of a routine interval chest film taken during the course of supervision of a "contact." These 3 individuals had lesions of minimal extent and were asymptomatic at the time of discovery.

During January, 1949 the Bureau of Tuberculosis of the Department of Health, New York City, conducted its *second* survey of the Municipal Lodging House, a city institution which offers shelter to homeless and destitute men. Two thousand five hundred and seventeen photofluororoentgenograms were taken of men who, for the most part were over 40 years of age, with the average age 56 years. Many of those examined had been included in the first Municipal Lodging House survey conducted in 1940–1941. Fortunately, the films of the original survey eight years ago were preserved on microfilm.

A detailed report of the findings of this recent survey will be published in a forthcoming paper. However, there are already available data which, although incomplete, are of sufficient interest to warrant inclusion here. Nineteen men, with normal survey films in 1940–1941 (as determined by reviewing the microfilms of the original survey roentgenograms) had roentgenographic evidence in their recent examinations highly suggestive of clinically significant pulmonary tuberculosis. *Sixteen of these cases were over 40 years of age at the time of the normal survey film, eight years ago.*

The detailed examination of these cases is still incomplete, although all of the patients have had 14 by 17 films taken. Sputum studies on most of the cases, however, are not as yet available. On the basis of the roentgenograms alone, therefore, the following preliminary diagnoses were made in these 16 cases:

Active far advanced pulmonary tuberculosis in 3 cases.

Active moderately advanced pulmonary tuberculosis in one case.

Minimal to moderately advanced pulmonary tuberculosis of undetermined activity in 11 cases.

Arrested minimal pulmonary tuberculosis in one case.

Acknowledgment

The author is deeply grateful to Dr. David Reisner, Denver, Colorado, for giving him permission to review his personal records on the 17 cases included in his study on incipient pulmonary tuberculosis.

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TUBERCULOSIS IN THE GERMAN POPULATION, UNITED STATES ZONE OF GERMANY[1]

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INTRODUCTION

The rise in tuberculosis mortality which accompanied World War I in all the belligerent countries of Europe gave promise of a similar rise after the last war, with its even greater devastation and longer duration. Such a rise did occur and was proportionately though not absolutely greater than that in World War I.

During the past year a number of reports have appeared dealing with the current tuberculosis situation in Germany. From certain of these reports, and from other sources, an impression has been formed not only that there has been a disastrous increase in the disease but that it is continuing to increase. Such opinions are to a large extent based upon the increase in reported tuberculosis morbidity. Other studies, which have placed more emphasis on mortality data, have come to different conclusions.

A survey of the postwar situation in the United States Zone of Germany, conducted by the authors in February, 1948 as members of a Commission of the Department of the Army, had as one of its objectives the collection of information on the extent of the rise in tuberculosis and the immediate postwar trend in that area. In view of the conflicting opinions mentioned above, it is believed worth while to present the evidence which was obtained. The data will be discussed under two main headings: first, mortality, with consideration of changes by age, sex, and form of disease; and second, morbidity, with discussion of factors responsible for apparent changes in morbidity rates. Brief consideration will also be given to the present program for tuberculosis control.

Mortality

In analyzing the present problem, it is pertinent to consider the rise in mortality which accompanied World War I. This has been reviewed by Drolet (8) who gives data showing that of 24 countries, mostly in Europe, all but 5 had a wartime rise. In 3 of these countries the highest rate was reached in 1916; in 3 it came in 1917, and in 13 it came in 1918. The influenza epidemic of that year

[1] This paper is based upon the findings of a Commission appointed by the Secretary of the Department of the Army and organized by Mr. Tracy S. Voorhees, Assistant to the Secretary, and Col. Tom F. Whayne, MC, Chief of the Preventive Medicine Division, Office of The Surgeon General. The members of the Commission were Doctor Long, Doctor Sartwell, Col. Silas B. Hays, and Maj. Alonzo W. Clark. In Germany the group was joined by Lt. Col. Moseley.

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undoubtedly caused the 1918 rates to be higher than they would otherwise have been. In none of the 24 countries was there a postwar rise between 1918 and 1920, although in several the 1920 rate remained higher than that for 1913. The promptness with which tuberculosis mortality responded to war and postwar changes is remarkable. How much of the rise resulted from the deaths of persons already suffering from active tuberculosis and how much from new infections or reactivation of old disease cannot be determined.

In Germany the mortality rate, which had been declining for a number of years, began to rise in 1915, was highest in 1918, and then fell off sharply to a point in 1920 which was just above the prewar level. With the disastrous inflation and depression of 1922–1923 a second but less marked rise occurred, succeeded by a further steady decline up to the beginning of World War II. In 1939 only four countries, Denmark, Australia, the Netherlands and the United States, had mortality rates lower than Germany's rate of 50 per 100,000.

Rather promptly at the beginning of World War II tuberculosis mortality began to rise in most of the belligerent European countries (2, 3, 6). The worst situations developed in Poland, Yugoslavia, Greece and Austria. On the other hand, Switzerland, Sweden and Denmark, countries which were not directly involved in the fighting, escaped any appreciable rise. The peak year of mortality was 1941 in the cities of London, Brussels and Paris; 1942 in Prague; 1944 in Rome and Warsaw; and 1945 in Amsterdam and Vienna. Daniels states that "the number of additional deaths from tuberculosis during the war must run into many hundreds of thousands, and the number of surviving sufferers must number between 5 and 10 million." It is, however, encouraging to note that in nearly all the war-torn countries tuberculosis as measured by mortality is again on the decline, and that in a considerable number of these countries it is already below the prewar level (6).

At the time of an official visit by one of the authors to Germany in October, 1945 (10), several adverse factors were noted which created a most grave situation. These included: a shortage of beds (less than one bed for tuberculosis per annual death), and a large number of open cases living at home; a housing shortage (density of population 1.5 to 2.5 persons per room in the large cities); nutritional difficulties; and the fact that before the close of hostilities patients with open tuberculosis had been generally permitted to work in factories and other industrial plants. This last factor was thought to have led to a spread of the disease which would become evident with the passage of time. A second visit in August, 1947 (11) revealed marked improvement in the bed situation and other tuberculosis control measures and permitted a more optimistic view.

The difficulties in the way of obtaining accurate mortality data from Germany for the war and postwar periods are great. In the first place, there have been mass migrations since the war which have made it very difficult to estimate populations. Large numbers of German prisoners of war (Wehrmacht) have been repatriated; there has been a great influx of displaced persons; and there has been a large immigration of German national expellees from Czechoslovakia and Poland. These immigrations have produced a considerable net increase

in the population of the U. S. Zone. The increase from 1939 to October, 1946, when a census was taken, was about 2.5 million, or about 18 per cent of the 1939 population, and was due chiefly to postwar immigration rather than to natural increase (12). There was a large excess of females, especially in the age span 20 to 29, between which ages in October, 1946 the ratio was 17 females to every 10 males.

Data on tuberculosis mortality are also subject to several errors. While official death registration figures have been used almost entirely, it should be noted that in some areas death certificates have until recently been signed by laymen. The changes in boundaries of the Länder or provinces within the U. S. Zone have rendered comparison of current mortality with earlier data impossible except in the case of Bavaria, which has remained largely unchanged in territory. Members of the German military forces are understood to be excluded from both population and mortality statistics from 1939 to the end of the war. The inclusion

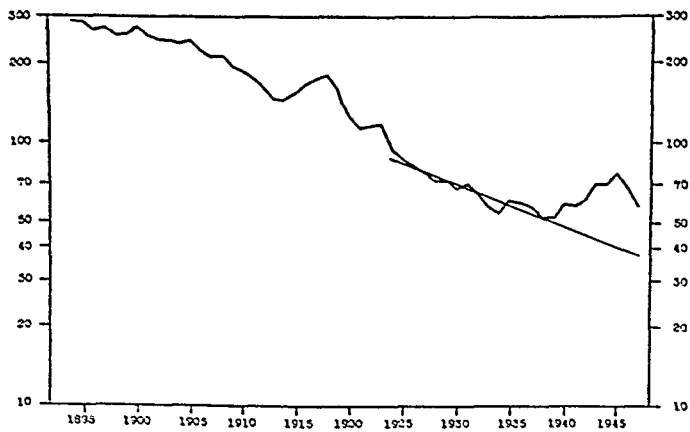

FIG. 1. Mortality rates for pulmonary tuberculosis, Bavaria, by year, 1894–1947

of displaced persons in mortality statistics is uncertain during a part of the postwar period. Finally it must be remembered that the disruption of German civil organization in the latter part of the war and immediate postwar period has undoubtedly reduced the accuracy of all statistics for this period; in fact, for some places such data are wholly lacking.

In spite of these deficiencies, it is fairly clear that tuberculosis mortality began to rise in most parts of western Germany in 1939 or 1940 and continued upward until about 1945. Since then it has been on the decline. The extent of the rise in western Germany was rather small by comparison with other parts of southeastern and central Europe. In Berlin, however, where the prewar rate was somewhat higher than elsewhere in Germany, the wartime rise was very large.

The statistics for Bavaria, which have been recently published (13, 15) will be examined in detail. Bavaria is a largely agricultural state in the south of Germany, its principal city being Munich. Little change was made in its territorial limits after the war. In figure 1 may be seen the trend of pulmonary tuberculosis mortality in Bavaria from 1894 to 1947, the scale of rates being logarithmic.

The trend line drawn on this graph is fitted by the least squares method employing the logarithms of the rates from 1924 to 1938. It is seen that both wars produced very similar interruptions in the long time downward trend of mortality, the rise in World War II being proportionately greater owing to the longer duration of hostilities and the other factors already mentioned. It is also apparent that the postwar decline has thus far been like that following World War I.

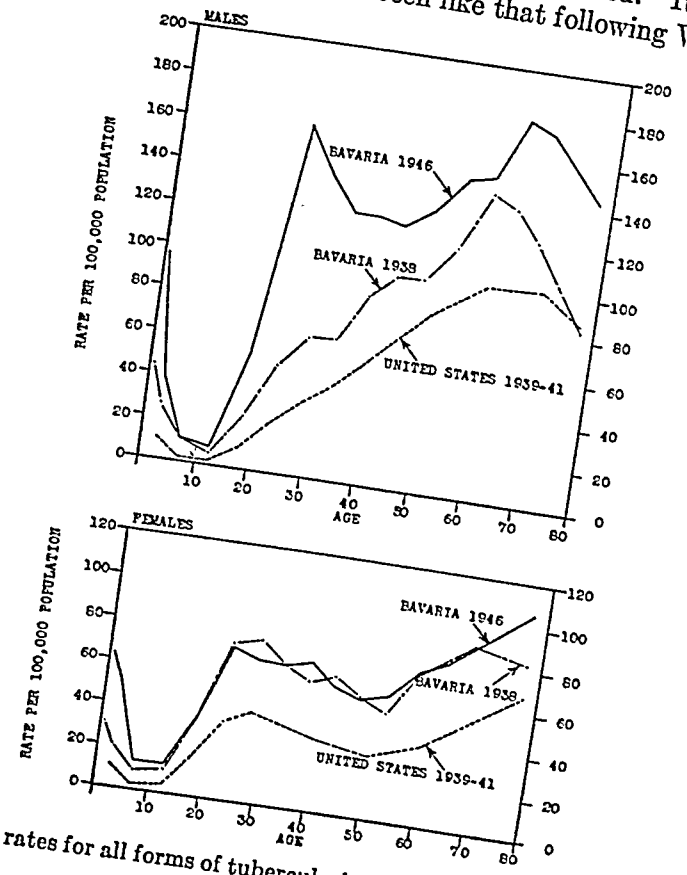

FIG. 2. Mortality rates for all forms of tuberculosis by sex and age, Bavaria and United States.

An attempt was made to obtain age- and sex-specific mortality rates for Bavaria, in order to show what population group was chiefly responsible for the excess mortality. The difficulty was not so much in the enumeration of deaths as in relating them to the population by age groups. It is emphasized that, owing to the extensive migrations mentioned above, the accuracy of these rates is open to question. They are presented in figure 2, however, as giving some indication of where the excess lies. In this graph (figure 2), age-specific rates for each sex are shown for the years 1938 and 1946, with the corresponding rates for the white population of the United States averaged for 1939–1941 (the age groups being slightly different).

In 1938, Bavaria's age distribution of mortality in both sexes was roughly

parallel to, although somewhat higher than, that in the United States. In 1946 the mortality for males was greatly increased in infancy and in all adult age groups. The most marked excess was in the 20 to 34 year age span. The opinion was expressed by German health officers that excessive mortality among repatriated prisoners has contributed to this high rate. Roentgen surveys of such groups have revealed a high prevalence of significant tuberculosis.

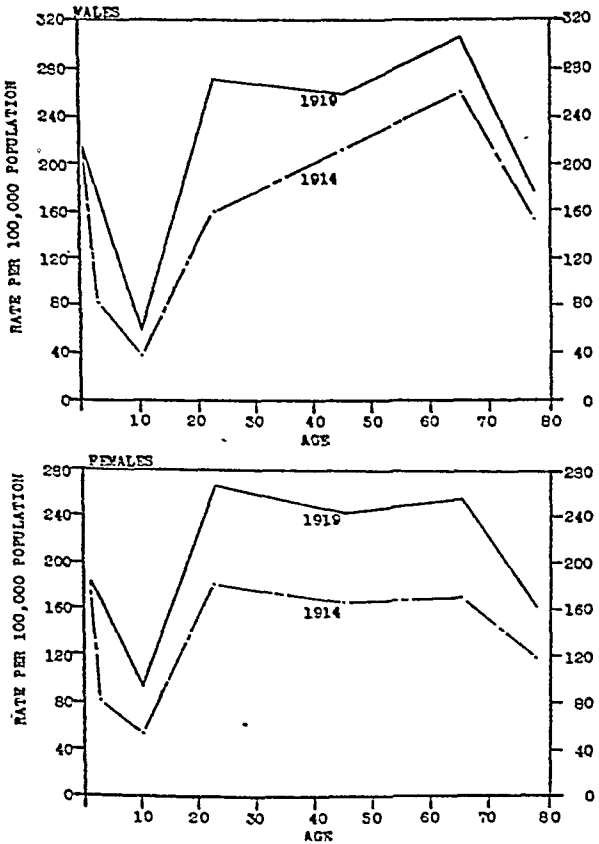

FIG. 3. Mortality rates for all forms of tuberculosis by sex and age, Germany, 1914 and 1919.

Among females, on the other hand, the only population groups showing an increase in 1946 as compared with 1938 were infants and young children, among whom a rise similar to the male rise was recorded. There was practically no difference between prewar and postwar mortality in adult females. Presumably had there been no war, the 1946 rates would have shown a decrease, but at least there was no rise. The increase in infant mortality from tuberculosis in both sexes suggests that, in this age group at least, the frequency of infection was greater in 1946 than before the war.

For comparative purposes, death rates for all Germany for the years 1914 and 1919 were obtained (14) and are shown in figure 3. It will be noted that in World War I the increase in male mortality was much the same as in World

War II; but female mortality also increased at all ages in about the same way as male mortality, in contrast to what happened in Bavaria in World War II.

The other Länder in the U. S. Zone have experienced rises and declines in tuberculosis mortality quite similar to that of Bavaria. The evidence indicates that mortality was highest in 1945 and has steadily fallen since that year. Indications of this are seen in figure 4 in which the trend of tuberculosis mortality

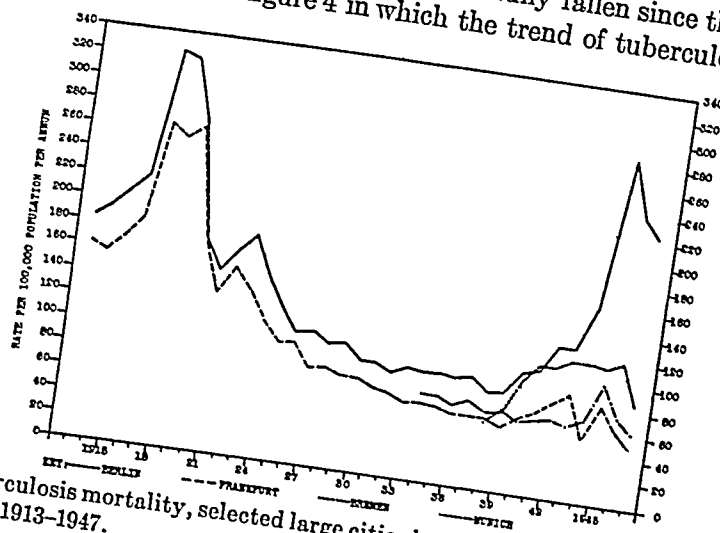

FIG. 4. Tuberculosis mortality, selected large cities in Germany, death rates for all forms of tuberculosis, 1913–1947.

TABLE 1

*Mortality Rates per 100,000 Population for all Forms of Tuberculosis, U. S. Zone of Germany and Berlin, 1946 and 1947**

	MORTALITY RATE	
	1946	1947
Land Bavaria	75	67
Land Hesse	72	69
Land Württemberg-Baden	77	67
Bremen (city only)	119	87
Berlin (entire city)	239	223

* See text for comments on reliability of rates.

rates for four German cities, all of them except Berlin being in the U. S. Zone, are depicted, and in table 1, which gives rates for 1946 and 1947 in each part of the U. S. Zone.

Land Hesse shows an excess in mortality of adult males from age 20 upward. This excess is noted both by comparison with female rates and with prewar male rates. Thus, despite the considerably larger number of females in the population, there were 1,733 male deaths from tuberculosis in 1947 and only 1,136 female deaths. Among males, tuberculosis accounted for 7 per cent of all deaths; among females, for 5 per cent. In Frankfort, the largest city of Land Hesse, the

data furnished indicated that the tuberculosis mortality rate by 1947 had already fallen below the prewar level despite the fact that Frankfort suffered heavily by bombing and the resultant overcrowding. Frankfort's increase in mortality was mostly limited to adult males and especially males over the age of 50, there being little change in female rates between 1938 and 1946.

Land Württemberg-Baden in 1947 reported 1,443 male deaths and 1,046 female deaths from tuberculosis, the excess among males being chiefly from age 30 upwards. Among males 6.8 per cent of all deaths were from tuberculosis while the corresponding figure for females was 5.2 per cent.

The territory comprising Land Bremen, a small area on the North Sea, has a population of only 494,000, of whom 395,000 are in the city of Bremen proper. In Land Bremen, the excess of male tuberculosis deaths is not apparent except above the age of 55. In 1947 at all ages there were 209 male tuberculosis deaths

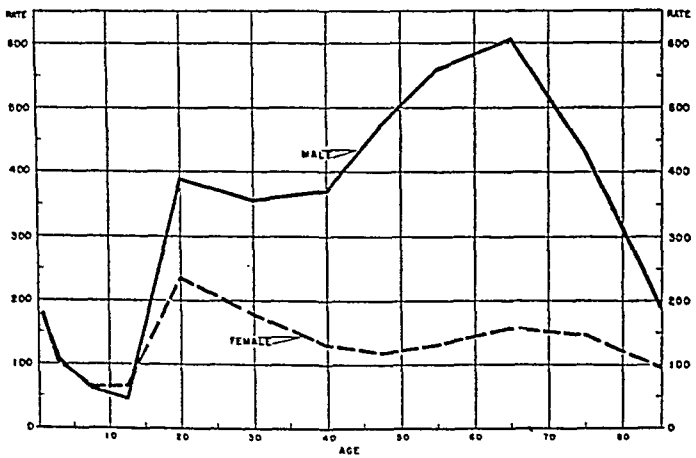

FIG. 5. Death rates for all forms of tuberculosis per 100,000 population, by sex and age, Berlin, 1946.

as against 173 female; the proportion of all deaths ascribed to tuberculosis was 7.4 for males and 7.3 for females.

The tuberculosis experience of Berlin was quite unlike that of the cities in the Western Zone of Germany and more closely resembled that of the cities of eastern Europe, where the rise was catastrophic. The data to be quoted refer to the entire city, not merely the U. S. sector. The trend of mortality has been seen in figure 4. The age distribution of death rates again shows a great excess of adult males over adult females, which is most pronounced over the age of 50 (figure 5). From age 30 upward there were 3,953 male tuberculosis deaths in Berlin in 1947 as against 1,737 female. Why Berlin suffered so heavily is not entirely clear. Certain reasons may, however, be advanced. Berlin had a higher rate than other large German cities before the war; bombing was continuous rather than sporadic; food shortages were presumably greater because of the difficulty of a large city in procuring food from the surrounding farmland; and Berlin was cut off from the use of its sanatoriums, most of which were outside the

city, to a greater degree than other places. Berlin's population is older than in other parts of Germany; with the present age distribution of deaths, this in itself would tend to make the crude death rate somewhat higher.

The relative frequency of pulmonary and other forms of tuberculosis as causes of death showed no remarkable variation in the areas studied, pulmonary tuberculosis ranging from 91 per cent of all tuberculosis deaths in Berlin down to 82 per cent in Hesse in 1947. For the three principal Länder in the U. S. Zone, 85 per cent of the mortality was ascribed to pulmonary tuberculosis and 15 per cent to tuberculosis of other organs. These proportions are identical with the figures for all Germany over the three year period, 1935–1937.

The proportionate mortality or percentage of all deaths ascribed to tuberculosis, in the three principal Länder of the U. S. Zone averaged 5.8 per cent in 1947. The proportionate mortality for Germany in the period 1935–1937 was 6.0 per cent. The comparable figure for the United States in 1945 (all races) was 3.8 per cent, and for the white race 3.1 per cent.

Morbidity

Reports that tuberculosis is epidemic and uncontrolled in Germany have been largely based upon the rise in morbidity reported by health departments. It has been predicted that this rise must be followed after a time by a rise in mortality. Morbidity figures from all the Länder have been carefully studied, and it was the opinion of the Commision that they probably do not reflect a real increase in tuberculosis.

In the United States relatively little attention is paid to the trend in reported cases of tuberculosis, except as an index of case finding. Case reporting is used primarily for getting the patient and his family under health department supervision, and for assisting in the provision of sanatorium care for the patient and examination of his contacts. Ordinarily too many variables affect the recognition and reporting of the disease to make comparisons of morbidity at different times and in different places meaningful. Most of these variables are operative in Germany, and in addition there are several not encountered in the United States.

The classification of active tuberculosis cases presently employed in German public health practice is rather elaborate, having three categories of respiratory tuberculosis and two categories of nonpulmonary tuberculosis. The three respiratory categories may be briefly described as:

(a) Active respiratory tuberculosis, with demonstrable tubercle bacilli.

(b) Active respiratory tuberculosis, without the demonstration of tubercle bacilli but with lesions which are regarded as "open" or infectious. Terms sometimes used for this group are "clinically open" or "facultative open".

(c) Active "closed" or "noninfectious" respiratory tuberculosis. This group includes stationary infiltrations, hilar and bronchial node enlargements, miliary strand-like lesions of lungs, exudative pleurisy, productive cirrhotic lesions, and infants with positive tuberculin reactions.

Most of the types of disease in group (c) would not ordinarily be reported in

the United States. In the absence of knowledge of how large a proportion they constitute, it is felt that this group is best excluded from consideration in the analysis of morbidity.

Morbidity figures from Germany are based upon reports rendered by the tuberculosis clinics or Fürsorgestellen, in which nearly all known cases are registered and where most of the diagnoses are checked and confirmed. In this respect they are more reliable than American data. There are, however, two important factors operating to cause an apparent increase in morbidity. The first of these is the fact that tuberculosis cases are authorized to receive supplementary food rations. This would appear to create an incentive for physicians to report borderline cases and for patients to seek medical care.

The second, and more important factor, is the greatly increased employment of case finding techniques. During and immediately after the war the Fürsor-

TABLE 2A

Initial Examinations and Diagnoses in Bavarian Fürsorgestellen in 1947, by Quarters

PERIOD	INITIAL EXAMINATIONS MADE	NEW CASES OF ACTIVE PULMONARY TUBERCULOSIS REPORTED		CASES PER 100 EXAMINATIONS	
		"Open"	"Closed"	"Open"	"Closed"
January to March	40,277	1,663	4,367	4.1	10.8
April to June	56,684	2,258	6,624	4.0	11.7
July to September	61,437	1,937	5,781	3.2	9.4
October to December	58,956	1,618	5,940	2.7	10.1

Note: "New cases" does not include those cases previously registered with the Fürsorgestellen as "closed" or inactive tuberculosis and subsequently transferred to the above groups. Such cases constitute about two-fifths of the total "active open" and one-fifth of the "active closed" cases reported.

gestellen were unable to operate effectively; but during 1946 and 1947 their case finding activities have been greatly intensified. Fluoroscopy is very widely used in Germany as a diagnostic procedure, the use of roentgenograms being often reserved for cases in which fluoroscopic examination has given positive or suspicious findings, and German physicians are highly skilled in this technique. Records are kept of the number of fluoroscopic examinations per month made by the Fürsorgestellen, and the use of the procedure has increased greatly during the past two years. The larger the number of persons examined, the higher will be the yield of new cases of tuberculosis, although the yield per 100 examinations may diminish. This is illustrated by table 2A in which the number of initial clinic examinations in all the Fürsorgestellen of Bavaria during 1947 is related to the total number of new cases of open and closed active pulmonary tuberculosis reported by the same agencies. While the number of examinations increased up to the third quarter, the number of new cases identified failed to increase proportionately, and in fact there was a definite downward trend over the year in the yield of "open" cases in relation to the number of examinations for tubercu-

losis. Table 2B gives data from the same source for an eighteen month period ending in June, 1948, showing total examinations and total cases of open tuberculosis.

In most of the other areas studied it can be shown that such increases in new cases as have occurred have been in "closed" cases (see category (c) above). One cannot avoid looking with some skepticism on this increase, which means either that a change is occurring in the character of the tuberculosis cases which come to light, or that something is happening to bring the "closed" cases to light relatively more often than formerly. Possible explanations for such a rise are the

TABLE 2B

Total Examinations in Bavarian Fürsorgestellen and Cases of Open Tuberculosis, by Quarters, January 1947 to June 1948

PERIOD	TOTAL EXAMINATIONS MADE	"OPEN" PULMONARY TUBERCULOSIS CASES REPORTED*	CASES PER 100 EXAMINATIONS
January to March, 1947	81,180	2,706	3.3
April to June, 1947	109,052	3,508	3.2
July to September, 1947	112,994	3,152	2.8
October to December, 1947	112,294	2,813	2.5
January to March, 1948	128,341	3,140	2.4
April to June 1948	136,923	3,565	2.6

* Includes all cases of bacteriologically or clinically "open" pulmonary tuberculosis diagnosed during the quarter.

TABLE 3

Total Cases of Tuberculosis Registered at Fürsorgestellen in Berlin in 1947, by Quarters.

	ACTIVE "OPEN" PULMONARY BACT. POS.	BACT. NEG.	ACTIVE "CLOSED" PULMONARY	EXTRAPULMONARY
January, 1947	11,895	9,185	43,850	6,433
April	11,795	9,626	46,980	7,156
July	11,609	9,768	48,559	7,842
October	11,820	9,972	51,272	8,431
January, 1948	11,241	9,769	49,188	8,707

improvements in diagnostic techniques and the incentive to report this type of case.

Another measure of the tuberculosis problem, which also is not employed as such in this country, is the prevalence of the disease as determined from the total number of cases registered with the health departments. Obviously, this is influenced not only by current case finding but also by the efficiency with which old cases are removed from the register after death, moving away, or recovery. It was not possible to ascertain how carefully the registers were maintained, although a very favorable impression was gained of the methodical and precise methods of record keeping and character of nursing supervision in the few clinics visited. The number of registered cases in Berlin by quarters over a period of one year ending in January, 1948 is shown in table 3. There has been an actual

decline in the number of bacteriologically positive pulmonary cases and an equivalent increase in bacteriologically negative "open" cases; a definite increase in the "closed" cases; and a rather marked increase in extrapulmonary tuberculosis. The reason for the latter increase is not known; it may perhaps, as some have suggested, indicate a real increase in chronic extrapulmonary forms which has not yet been reflected in a rising mortality. These figures, however, do not substantiate a real increase in clinically significant pulmonary tuberculosis.

Conditions in the British Zone of Germany have been analyzed in a report by Daniels and Hart (5). These workers obtained findings very similar to those which have been presented above and their conclusions are substantially the same as ours. So far as tuberculosis is concerned there is evidently little difference between conditions in the British and U. S. Zones.

Tuberculosis Control Program

Space does not permit a detailed discussion of the tuberculosis control program. In brief, with American assistance, bed provisions have been greatly increased until now the number of hospital and sanatorium beds per annual death, except in Berlin, approaches the present ratio in the United States. There are still, however, too few beds to meet the accepted standards, particularly in Berlin, and many open cases must be cared for at home. Local public health organizations appear to be doing an aggressive and effective job of case finding and case supervision. A BCG vaccination program has been undertaken under the supervision of the Danish Red Cross. Food supplements are authorized for tuberculous persons, which on the whole provide an adequate dietary for this group when the food can be obtained. Germany is greatly handicapped by an extreme shortage of many essential materials in addition to food, ranging from soap to X-ray film. Only general economic recovery can help this situation, and indeed the whole tuberculosis problem is closely linked to the economic state of the country.

SUMMARY AND CONCLUSIONS

1. Tuberculosis mortality in the U. S. Zone of Germany began to rise promptly at the beginning of World War II, reached a peak in 1945 and has progressively declined in 1946 and 1947. It is still above the prewar level in most places and is well above the rate to have been expected if the prewar downward trend had continued. The extent of the rise was only moderate as compared with that in several other European nations.

2. The increase in mortality appears to have affected infants of both sexes and, to an even greater degree, adult males. Females above the age of one year showed little increase as judged by age- and sex-specific rates for 1946. This is in contrast with the rise in World War I, in which both sexes and all age groups participated. The reason for the difference is not apparent.

3. Case finding programs have been steadily improved and intensified since the resumption of public health activities. This factor and others which have been discussed are believed to be responsible for the apparent postwar increase in tuberculosis incidence. No convincing evidence has been found for a real increase in the incidence of active open pulmonary tuberculosis over the past two

years. The outlook appears hopeful for continued reduction in tuberculosis, barring fresh economic or military disaster.

SUMARIO Y CONCLUSIONES

Tuberculosis en la Población Alemana de la Zona Estadounidense de Alemania

1. La mortalidad tuberculosa en la Zona Estadounidense de Alemania comenzó a elevarse poco después del comienzo de la Guerra Mundial II, alcanzó su acmé en 1943 y ha bajado gradualmente en 1946 y 1947. Todavía supera la cifra preguerra en la mayor parte de los sitios, hallándose bien por encima del coeficiente que hubiera cabido esperar, de haber continuado la tendencia descendente de antes de la guerra.

2. El aumento de la mortalidad parece haber afectado a las criaturas de ambos sexos, y aun más, a los varones adultos. Las hembras de más de un año revelaron poco aumento, a juzgar por los coeficientes específicos por edad y por sexo para 1946, lo cual contrasta con el aumento en la Guerra Mundial I, en el cual participaron ambos sexos y todos los grupos etarios. La causa de la diferencia no es evidente.

3. Las obras de descubrimiento de casos han sido mejoradas e intensificadas constantemente desde que se reanudaron los trabajos de sanidad. Estas obras y otros factores pasan por ser la causa del aparente aumento de la incidencia tuberculosa después de la guerra. No se han encontrado pruebas convincentes de un aumento real de la incidencia de la tuberculosis pulmonar abierta activa durante los dos últimos años. De no sobrevenir nuevos desastres económicos o militares, cabe esperar que continue la disminución de la tuberculosis.

Acknowledgment

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WHAT DOES A ROENTGENOGRAPHIC SURVEY TEACH THE PUBLIC?

Including a Comparison with Other Tuberculosis Information Polls

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(Received for publication November 10, 1948)

INTRODUCTION

Case finding is of vital importance in a tuberculosis control program. Mass roentgenographic surveys accompanied by a large scale educational program have become more and more the order of the day. The results of such projects in terms of numbers are easy to measure. Educational results are not. Frequently, when the numbers examined roentgenographically do not reach expectations, the statement will be made that, no matter what the numbers examined, the mass study was an effective health education campaign and the people learned a lot about tuberculosis. The Montgomery County Tuberculosis Association resolved to find out whether such is in fact the case.

A committee was formed, consisting of C. Mayhew Derryberry, Ph.D., Chief, Office of Health Education, U. S. Public Health Service; Mrs. Leona Culp, Special Consultant, Office of Health Education, U. S. Public Health Service; K. Frederick Welte, M.D., Director of Tuberculosis, Montgomery County Health Department; Mrs. Jane A. Shepherd, Director, Public Opinion Poll, Washington Post; Miss Claudia Galiher, Executive Secretary, Montgomery County Tuberculosis Association; and Miss Stella Randolph, Director, Public Health Education, Montgomery County Health Department. Technical advice on the sampling was obtained from Mr. Walter Perkins, Statistician with the Statistics and Analysis Division, Bureau of Public Assistance, Social Security Board.

The committee decided to poll a sample of the county two months in advance of roentgenographic survey and to repeat the same poll on a similar sample of the same area just two months after the conclusion of the survey. The Montgomery County chest roentgenographic survey was conducted simultaneously with the District of Columbia Survey, so that Montgomery County people were doubly barraged with information about tuberculosis and chest roentgenograms.

The first poll was conducted in October, 1947. The Montgomery County chest roentgenographic survey lasted from January to March, 1948, during which time two machines were in constant operation and an intensive educational campaign was put on. The second poll was accomplished in May, 1948.

An educational campaign was carried out, based on organizing the entire community to participate in the project. Plans were formulated in such a way as to involve as many groups and individuals as possible. Volunteer committees worked on over-all planning, locations and hours, a speaking program to reach all organized groups, the recruitment of volunteer clerical personnel to work with the roentgenographic units, distribution of posters, a house to house canvass, special roentgenographic arrangements for businesses. Newspapers, radio, thea-

ters, schools, and churches all played an important part in spreading information concerning the survey.

The objectives of the study were broadened from a measurement of a particular educational project to include the obtaining of information for program planning purposes, so that, as finally stated, the objectives were:

(*1*) To test the accomplishments of the educational program of the tuberculosis association in one campaign between October, 1947 and May, 1948.

(*2*) To provide information concerning people's knowledge of and attitude toward tuberculosis, which will be useful to the tuberculosis association in carrying out its educational program.

The Questionnaire

A questionnaire was developed by the committee named above. All the questions were designed for free responses on the part of the persons interviewed, in order not to suggest possible answers and thereby introduce bias into the study. For purposes of comparison a number of questions previously used in a Gallup Poll were incorporated. The questionnaire was tested and revised before being printed and put into use.

The questions covered the following areas: (*a*) the cause and communicability of tuberculosis; (*b*) the cure for tuberculosis; (*c*) symptoms in tuberculosis; (*d*) the value of roentgenographic examination, and for whom; (*e*) stigma attached to tuberculosis; (*f*) attitude toward people who have had or who have tuberculosis; (*g*) personal responsibility in the control of tuberculosis; (*h*) personal acquaintance with tuberculosis, especially in family situations; (*i*) exposure to tuberculosis education.

THE INTERVIEWERS

Volunteer interviewers were obtained from the area in which the poll was to be taken and came primarily from a group of women interested in and active in the local Health Center.[1] These women all attended a training session at which the purpose of the poll was carefully outlined, the selection of the sample explained, the composition of the questionnaire gone over, and interviewing technique and recording carefully explained. Sample interviews were presented for illustrative purposes. Interviewers were impressed with the importance of carrying out the instructions given them in order not to impair the significance of the study.

THE SAMPLE

Silver Spring, Maryland, was selected as the portion of Montgomery County in which the poll would be conducted, since it has the greatest concentration of people in the county and also because it is an area in which there is great economic differentiation.

The sample was carefully chosen on a household basis. This system was used to cut down the bias occurring when interviewers are allowed to choose their own respondents. Also, it automatically gives an accurate sample by economic status, as homes are chosen from all parts of town. The area to be polled was

[1] Silver Spring Public Health Lay Committee.

marked on a map, and each block in the area was assigned a number. Then one out of every four blocks was picked at random, using J. H. C. Tippett, Random Sampling numbers. Within each of the selected "random" blocks, every sixth household unit was taken, starting alternately at the north, east, south, and west corners. Thus, block 1 was used, representing the first four blocks, the first household, starting at the north corner; block 7 from the second four blocks, the second household, starting at the east corner; block 12 from the third four blocks, the third household from the south corner, et cetera. Three hundred five addresses were obtained in this manner for the first sample. The second sample of 332 addresses was obtained in a similar manner. The blocks used in the first sample were excluded from the second, so that the opinion poll itself would not

TABLE 1

Composition of Samples

	POLL 1 (297 INTERVIEWS)	POLL 2 (326 INTERVIEWS)
	Per cent	Per cent
Total interviews	100	100
Both sexes	100	100
Male	30	100
Female	70	32
All ages	100	68
Under 30	33	100
30 to 44	44	29
45 and over	22	46
Unknown	1	24
All educational levels	100	1
School	13	100
High school	50	9
College	35	52
Unknown	2	38
		1

affect the results. Volunteers were given specific household addresses. Each volunteer had approximately 10 interviews.

Substitutions were discouraged and specific instructions were issued as to how they should be made, if necessary. Approximately the same number of men and women were to be interviewed. The lowest age acceptable was 15 years, the same group as was eligible for a chest roentgenogram in the survey. The age of the respondents was to be recorded on the schedule according to the guessed observation of the interviewer.

Two hundred ninety-seven (297) interviews were obtained in sample 1, and 326 in sample 2. The difference between the number of addresses obtained and the number of interviews made is a consequence of refusals and the fact that in some areas homes had been vacated to make room for the expanding business section of the town.

The age and educational distribution of the samples is believed to be similar to that of the population of Silver Spring but no accurate figures are available on

the composition of the population at this time. The 1940 census figures on the population of Montgomery County show an older and a less highly educated group. There is reason to believe, however, that Silver Spring, a city that is largely a suburb of Washington, D. C., and which grew rapidly during the war, is made up of a younger, better educated group than that reported in 1940 for the whole of Montgomery County. The sex distribution in the sample is definitely at variance with the existing population. It is likely that there are slightly more women than men in the area, but the preponderance of female interviews is probably a result of the fact that women were more readily available at home. This result was obtained in spite of instructions to interviewers to try to obtain half men and half women in their interviews. This has apparently made little difference to the sample, as the amount of knowledge held by men and women regarding tuberculosis was similar.

RESPONSES

A tabulation was made of the questions asked. Here the questions have been rearranged by subject matter for ease of presentation. In the interview the questions were carefully placed to minimize the influence of one question upon the answer to another.

The Cause and Communicability of Tuberculosis

The following questions were asked in order to determine what ideas people have as to the cause of tuberculosis and whether or not they think it is catching:

(*1*) *"What do you think is the cause of tuberculosis?"*

	Per cent of Responses	
	Poll 1	*Poll 2*
Germs	21	26
Undernourishment or malnutrition	25	30
Neglect of health	21	17
Run-down condition or low vitality	26	26
Living conditions	19	17
Heredity	12	8
Don't know	5	14
Other	12	8

There seems to be great confusion among the respondents with regard to the cause of tuberculosis. The so-called contributory factors, malnutrition, neglect of health, and run-down condition were indicated as often as germs. Recognition of the frequent accompanying factors of disease as a basis for social action is important in a broad community-prevention program. One-tenth of the respondents mistakenly named heredity as the cause. The responses add up to more than 100 per cent, as each person was allowed several responses.

There was a slight improvement in people's knowledge between the two samples. In the second poll five per cent more mentioned germs, 4 per cent less mentioned heredity. An interesting phenomenon occurred with the "don't know's." The percentage so responding was 5 per cent on sample 1 and 14 per cent on sample 2. It may be that persons having fixed erroneous ideas experience feelings of doubt when confronted with "facts."

(2) *"Do you think tuberculosis is catching?"*

	Per cent of Responses Poll 1	Poll 2
Yes	84	85
No	11	9
Don't know	5	6

In spite of confusion as to the specific cause of tuberculosis, the great majority of people realize that it is a contagious disease.

(3) *"Do you think a baby can be born with tuberculosis?"*

	Per cent of Responses Poll 1	Poll 2
Yes	36	34
No	42	40
Don't know	22	26

The situation regarding heredity and tuberculosis, as reflected in the first question, is not absolute, inasmuch as, when directly asked if a baby can be born with tuberculosis, over one-third replied "yes."

(4) *"How about yourself? In your opinion is it possible for you to get tuberculosis, or do you feel safe from this disease?"*

	Per cent of Responses Poll 1	Poll 2
Possible	68	76
Safe	31	22
Because older people safe	8	4
Recently or periodically checked	6	7
No tuberculosis in family	4	4
Am healthy	7	4
Other	6	3
Don't know	1	2

Generally speaking, people do realize that it is possible for them to get tuberculosis; 8 per cent more felt that way after the roentgenographic survey. There was a definite decrease in the percentage of older people who feel safe because of their age, a change which was not reflected in a change in the sample.

The Cure for Tuberculosis

The following questions were asked to determine whether or not people think tuberculosis is a curable disease, and what their ideas are with respect to the cure:

(1) *"Do you think a person who has tuberculosis can get well?"*

	Per cent of Responses Poll 1	Poll 2
Yes	95	95
No	4	2
Don't know	1	3

On both tests an overwhelming majority indicated that they thought tuberculosis was curable.

(2) *"What do you think a person who has tuberculosis should do to get well?"*

	Per cent of Responses Poll 1	Poll 2
Rest in bed	38	33
Good food	21	21
Proper medical care	61	67
Go to hospital	24	20
Fresh air	11	10
Proper climate	11	10
Other	7	4

The majority were aware of the importance of proper medical care. Rest in bed, good food, and sanatorium care were also recognized. Unfortunately, climate is still held as important in the cure of tuberculosis by 10 per cent of the people. It is probable that, had a direct question on climate been included, a larger percentage would have thought it important. These responses add to more than 100 per cent as multiple answers were permitted. Not one person on either poll indicated the use of drugs or medicine.

Symptoms in Tuberculosis

This question was asked to determine people's awareness of the absence of symptoms in early tuberculosis:

"In your opinion, when a person first gets tuberculosis, does he feel all right or does he have definite symptoms?"

	Per cent of Responses Poll 1	Poll 2
All right	46	48
Symptoms	43	40
Don't know	11	12

Less than one-half of the people questioned in each sample were aware that early tuberculosis may exist without symptoms. There was little change between poll 1 and poll 2, in spite of the fact that great emphasis was placed upon this point during the survey.

Age and the Occurrence of Tuberculosis

This question was asked to determine to what extent people were aware of tuberculosis as primarily an adult problem, and particularly a problem of the most productive years:

"As far as you know, at what age is tuberculosis most common?"

	Per cent of Responses Poll 1	Poll 2
Under 5	6	3
5 to 14	18	13
15 to 44	73	80
45 and over	9	7
Don't know	11	10

Responses to this question were recorded in terms of any or all age groups indi-

cated, so that the percentages add to more than 100. Most respondents were more aware of ages which fell, for recording purposes, in the 15 to 44 group than any other, *e.g.*, an answer of "the twenties" would be in this group. A lack of correct knowledge is shown by the number who indicated ages under 15 and by the very small number who indicated ages over 45.

The Value of Roentgenographic Examination

The following questions were designed to gather people's ideas regarding roentgenographic examination and its application:

(1) "*As far as you know, does the doctor have to have an X-ray before he can be sure that a person does not have tuberculosis or can he tell by a physical examination?*"

	Per cent of Responses	
	Poll 1	*Poll 2*
X-ray	69	68
Physical examination	12	10
Both	11	10
Don't know	8	12

About seven of ten persons think that roentgenographic examination is necessary before tuberculosis can be excluded. One of ten thinks that the doctor can tell by physical examination without a roentgenogram. Those who answered "both" were probably avoiding the issue, and fall for the most part into "don't know."

(2) "*Which is more important, would you say, for parents to be X-rayed, or for their young children to be X-rayed?*"

	Per cent of Responses	
	Poll 1	*Poll 2*
Parents	31	49
Children	26	13
Both	41	36
Don't know	2	2

There was a marked improvement on this question. This was one of the most stressed points during the survey. Emphasis was placed on the fact that only persons 15 years of age or over were to have roentgenograms. Some children were even turned away from the machines.

(3) "*Do you personally feel it important for everyone to be X-rayed?*"

	Per cent of Responses	
	Poll 1	*Poll 2*
Yes	81	96
Once a year	39	51
Periodically, but less often	25	27
As often as necessary	6	11
Don't know	11	7
No	15	3
Don't know	4	1

The survey undoubtedly convinced many people of the importance of roentgenographic examination. The per cent so responding jumped from 81 on poll 1 to 96 on poll 2. Approximately half the people on poll 2 thought people should be X-rayed once a year.

(4) *"Have you ever had an X-ray of your chest?"*

	Per cent of Responses Poll 1	Poll 2
Yes	64	89
Within year	20	65
1 to 5 years ago	28	18
Over 5 years ago	13	5
Don't know	3	1
No	36	11

This question might be considered to give an index to the response of the community to the roentgenographic survey. The above findings indicate that 89 per cent of the people in Silver Spring have had chest roentgenograms, all but 5 or 6 per cent within the last five years; 65 per cent (those who have been examined within the last year) were probably examined roentgenographically in the course of the survey.

Social Stigma of Tuberculosis

This question was designed to determine to what extent people feel that tuberculosis is something of which to be ashamed:

"If someone in your family had tuberculosis, would you feel free to talk about it or would you want to keep it to yourself?"

	Per cent of Responses Poll 1	Poll 2
Talk	80	80
Keep to self	15	15
No one else's business	5	7
Fear of social stigma	4	3
Shunning of contagious individual	4	4
Other	2	1
Don't know	5	5

It is interesting to note that there is no change in people's ideas regarding how freely they would talk about someone in their family having tuberculosis. Fifteen per cent of the people would keep such information to themselves and 5 per cent don't know whether they would or not. One person in thirty still feels a stigma attached to tuberculosis. The others who wouldn't talk gave reasons such as "no one else's business," "a matter between the individual and his doctor," and a fear of "shunning of the individual" because of the contagious nature of the disease.

Attitude toward People with Arrested Tuberculosis and Active Tuberculosis

The following questions were asked to determine people's attitude toward associating with persons in these two categories:

(1) *"Suppose one of the people you worked with, you learned one day, has spent some months in a hospital with tuberculosis but has been released by the doctor and is able to work. What would you think about this?"*

	Per cent of Responses	
	Poll 1	Poll 2
Situation all right	84	82
Situation bad but would do nothing	3	5
Sufficiently disturbed to check up on self or urge that something be done about the situation	13	13

Most people would feel no danger attached to associating with people who have been cured. However, one person in eight would react unfavorably.

(2) *"Suppose you had a job and you suddenly found out you were working right alongside a person who actually has tuberculosis. What would you do in this situation?"*

	Per cent of Responses	
	Poll 1	Poll 2
Nothing	10	7
Speak to employee	22	25
Report to employer	24	25
Avoid, or change jobs	23	26
Have check-up	22	23
Other	13	10

Most people would take action of some kind to improve the situation when closely associated in a working relationship with someone who has tuberculosis. However, the 7 per cent on poll 2 who would do nothing is still far too large.

Personal Responsibility in the Control of Tuberculosis

The following questions were included in the questionnaire to gather information as to the respondents' feeling of responsibility in a community-control program.

(1) *"A man you know had a chest X-ray at his office. He was sure he was in good condition. Later he received first one and then another letter from the Health Department asking him to come see the doctor. He became very much annoyed and told his friends that his health was none of the Health Department's business and that he was not going to see their doctor. What do you think of his course of action?"*

	Per cent of Responses	
	Poll 1	Poll 2
His attitude bad	94	97
His attitude all right	6	3

In this question a situation was presented in which the person concerned might or might not have tuberculosis, but is uncooperative. It was hoped that the responses would indicate an individual's own response to a similar situation. The majority felt that his attitude is definitely undesirable, with a slight improvement between polls. It may be that the small per cent who concur in his action would be uncooperative if patients themselves.

(2) *"Suppose a close friend of yours came to you and told you that for no definite reason she was afraid she might have tuberculosis. What would you advise her to do?"*

	Per cent of Responses	
	Poll 1	Poll 2
Have medical check-up or X-ray	96	96
Nothing	4	4

This question was designed, not only to discover what course of action an individual would advise for a friend who thought she might have tuberculosis, but in the hope that a conclusion might be drawn as to what he might do himself. An overwhelming majority would seek medical advice or have a chest roentgenogram, and no change was reflected between the polls.

(3) *"In chest X-ray surveys about 60 per cent of the people come and have an X-ray. What do you think is the main reason why the other 40 per cent do not?"*

	Per cent of Responses	
	Poll 1	*Poll 2*
Neglect or carelessness	48	61
Fear	25	24
Think they are all right	27	16
Don't know	4	4
Other	12	9

This question was designed to gather information as to reasons why roentgenographic surveys are not 100 per cent successful. It was hoped to determine from these responses to what degree people stay away from survey machines from fear. One-fourth of the respondents seem to feel fear a pertient factor. The main reason, however, is neglect or carelessness. There was a slight shift from "think they're all right," which might reflect an opinion that that is not a good reason for staying away.

(4) *"If an X-ray survey were to take place here, would you go and have a chest X-ray?"*

	Per cent of Responses	
	Poll 1	*Poll 2*
Yes	82	83
No	18	17
Am checked periodically	6	7
Physically unable to go	1	1
Not in favor	1	1
Too old to be worth while	3	2
Feel fine	4	1
Other	3	5

The results obtained through questioning respondents with regard to participation in a proposed roentgenographic survey were approximately the same on both polls, as were the reasons for not participating. Comparing this figure to the 96 per cent who think it important for everyone to be examined roentgenographically, it would seem that some of the 96 per cent feel that everyone but they should be so examined.

(5) *"Do you think that tuberculosis is an important problem in this community?"*

	Per cent of Responses	
	Poll 1	*Poll 2*
Yes	76	82
No	13	11
Don't know	11	7

This question was designed as a warm-up question, and was the first asked in each interview. Apparently, having a roentgenographic survey was sufficient proof for some that tuberculosis was important.

Personal Acquaintance with Tuberculosis

This question was asked to determine what percentage of the people have tuberculosis in their families or in close friends. It was hoped to find out whether the knowledge and attitude of those with tuberculosis in their families differed from that of the rest of the sample.

(1) *"Has anyone in your immediate family or a close friend ever had tuberculosis?"*

	Per cent of Responses	
	Poll 1	*Poll 2*
Immediate family	21	18
Close friend	17	16

One of every five respondents reported the existence of tuberculosis in their families at some time. It is possible that this figure might be even higher, for some of those with tuberculosis in their families might fall in the group which would "keep it to self."

SCORES

In order to compare the amount of knowledge of the various groups within the community and to evaluate the results of the educational campaign which took place in the course of the roentgenographic survey in Montgomery County, a system of scoring the tests was devised. The scoring is not intended to reflect the absolute level of knowledge held by Silver Spring. Another questionnaire or another scoring method would yield different results. The same method of scoring was used in both polls. The scores, by sex, on the two polls were as follows:

	Average Score	
	Poll 1	*Poll 2*
Both sexes	72.0	74.3
Male	72.2	72.4
Female	71.9	75.2

From the above it can be noted that there is only a slight increase in the average scores of poll 2 over poll 1, not more than might be attributable to a change in sample. It can further be noted that, while the degree of knowledge held by men and women appears the same on poll 1, there is a slight difference in favor of the females on poll 2 and that, if there has been any increase in knowledge, it is among the women.

The results were examined in terms of age groups:

	Average Score	
	Poll 1	*Poll 2*
All age groups	72.0	74.3
Under 30	75.5	74.7
30 to 44	73.5	76.2
45 and over	63.5	70.1

In the first sample the greatest amount of knowledge was held by persons under 30; the oldest age group was well below the average. In the second poll the 30 to 44 group were best informed, and the older people again lag. It is in the

45-and-over age group, however, that the greatest improvement between the polls occurs, the 30 to 44 group showing the next best improvement. The score of the under 30's has dropped slightly, but the difference is so small that it is almost certainly due to a difference in the sample.

Classified by the level of education, the results were:

	Average Score	
	Poll 1	*Poll 2*
All groups	72.0	74.3
School	63.6	71.5
High school	70.9	72.5
College	77.3	77.7

The knowledge regarding tuberculosis varied directly with the amount of education. Since tuberculosis education is not normally a part of elementary school education, however, and is usually given at high school and college levels, one might expect even more disparity. It would appear that, although knowledge regarding tuberculosis increases with education, other sources of information would appear to be at least equally important. Comparing the scores attained by the least educated group on the two polls, it is seen that the greatest improvement in knowledge occurred in this educational group. Perhaps this group recognizes that it does not know and is willing to learn.

If experience is the best teacher, one would expect people from families in which there has been tuberculosis to know more than the average person.

	Average Score	
	Poll 1	*Poll 2*
All groups	72.0	74.3
Those with tuberculosis in family	70.5	76.4
Those with no tuberculosis in family	72.3	73.8

Before the roentgenographic survey, the people with tuberculosis in their families were slightly below the average. After the survey, however, they were well above the average, their score having jumped 6 per cent, as compared to about 1½ per cent for the people with no tuberculosis in their families. This would seem to indicate that people with tuberculosis in their families, who have reason to be especially interested, learn more than the average when confronted with the opportunity to find out the true facts.

EXPOSURE TO TUBERCULOSIS EDUCATION

The respondents were asked the following series of questions to determine to what educational media regarding tuberculosis they had been exposed and to attempt to discover their relative values. The following table shows the per cent of the people exposed to the various media.

	Per cent of Responses	
	Poll 1	*Poll 2*
Have you ever read about tuberculosis in the newspaper? Yes	90	96
Have you ever seen any pamphlets or posters on tuberculosis? Yes	82	91
Have you ever heard anything about tuberculosis on the radio? Yes	77	86
Have you ever seen a moving picture on tuberculosis? Yes	34	43
Have you ever heard anyone talk or speak at a meeting on tuberculosis? Yes	27	37

During the course of the survey more people were reached with information on tuberculosis than had been reached before. The importance of the various media with respect to coverage was in this order: newspaper, pamphlets or posters, radio, moving pictures, speakers.

An evaluation of the various media was difficult to accomplish, as most people were exposed to several sources of information, and quite a few to all. However, consideration of the accomplishments of the various media in terms of scores attained on poll 2, shows the various media to be effective in this order: speakers, movies, radio, posters or pamphlets, newspapers. It would seem that the more personal approach is the more effective.

COMPARISON WITH OTHER POLLS

Public opinion polls are very popular today, not only for purposes of evaluation, but also as a part of basic planning. Some comparison with other polls recently completed would seem valuable.

During 1947, public opinion polls on tuberculosis were conducted by the American Institute of Public Opinion (Gallup Poll)[2] operating over the entire country, and in the following cities under various auspices, including tuberculosis associations: Hartford, Connecticut[3]; Denver, Colorado[4]; and Mishawaka, Indiana[5]. The questions used on the various polls are quite similar, and in some instances identical.

Comparable questions for the most part received similar answers in all of the polls. For example: "Do you think tuberculosis is catching?"

Polls	*Per cent answering "yes"*
Gallup	70
Hartford	80[6]
Denver	80[6]
Mishawaka	70
Silver Spring Poll 1	84
Silver Spring Poll 2	85

Silver Spring was somewhat better informed on this particular question than were the others reported above. This may be a consequence of the composition of the city, the citizens of which on the whole are well educated and high in economic status.

Questions on the possibility of inheriting tuberculosis were asked in all of the polls. *"Do you think a baby can be born with tuberculosis?"* or *"Do you believe that tuberculosis is inherited?"* brought the following responses:

[2] George Gallup, The Gallup Poll, as reported in the Washington Post, June 7, 1947, page 9.

[3] Chas. E. Lyght, Public Opinion Poll on Tuberculosis to aid Hartford Assn. in Revising Program, the NTA Bulletin, February, 1948, page 29.

[4] Chas. E. Lyght, Denver to Use Findings of Tuberculosis Poll to Strengthen and Extend Program, The NTA Bulletin, April, 1948, page 61.

[5] Cyril J. Hoyt, John Q. Public Attitudes affecting Tuberculosis Control, Monthly Bulletin, Indiana State Board of Health, June, 1948, page 128.

[6] The published figures are "8 out of 10."

Polls	*Per cent answering "no"*
Gallup	35
Hartford	30
Denver	40[7]
Mishawaka	43
Silver Spring Poll 1	42
Silver Spring Poll 2	40

People everywhere were poorly informed on this subject of the inheritability of tuberculosis.

A great discrepancy is seen in the answers to the question regarding the cause of tuberculosis. When asked *"What do you think is the cause of tuberculosis?"* as the third question in the Silver Spring poll (the first two questions were *"Do you think tuberculosis is an important problem in this community?"* and *"Why?"*) only 21 per cent on poll 1 and 26 per cent on poll 2 gave "germs" as the answer. The Gallup Poll results were similar, with a 22 per cent response of "germs." Reports from Hartford and Denver, however, indicate that at least two-thirds of their people knew that germs caused tuberculosis. It is interesting to consider why this difference exists. Is it that the people of Hartford and Denver are acutely aware of germs as the causative agent of tuberculosis? Was the response in some way connected with the way in which the question was worded or placed? Has the educational program in these two cities been so far above the country as a whole to make this difference? These are just a few questions in which the clue might be hidden. Different arrangement of questions might well bring different responses. For example, if this particular question were preceded by *"Do you think tuberculosis is catching?"* the interviewer might be considered to have inserted the idea of a causative agent in the respondent's mind.

Most people believe that a person can recover from tuberculosis, as seen by the percentages so reporting in the various polls:

Polls	*Per cent answering "cure possible"*
Gallup	83
Hartford	90[8]
Denver	75[9]
Silver Spring Polls 1 and 2	95

The citizens of Silver Spring appear to have a more optimistic outlook than the others.

Somewhat comparable questions were asked in Mishawaka and Silver Spring to determine people's attitudes toward the person with arrested tuberculosis. The responses indicate that 78 per cent in Mishawaka and 82 per cent in Silver Spring would be willing to work alongside a person who has recently recovered from tuberculosis. (Denverites generally believed that a person with arrested tuberculosis can resume normal living, only 19 per cent suggesting that an ex-tuberculosis patient would be under extreme restrictions.)

[7] Reported as "about four in ten."
[8] Reported as "9 out of 10."
[9] Reported as "3 out of 4."

On the question of whether or not a person would wish to keep to himself the fact that someone in his family had tuberculosis, reports are available only from Mishawaka and Silver Spring.

Polls	Per cent feeling free to talk
Mishawaka	82
Silver Spring Polls 1 and 2	80

"Feeling all right" as opposed to "symptoms in early tuberculosis" brought similar low responses from Mishawaka and Silver Spring.

Polls	Per cent answering "feeling all right"
Mishawaka	43
Silver Spring Poll 1	46
Silver Spring Poll 2	48

The degree of popular acceptance of the roentgenographic survey as an instrument in tuberculosis control might be measured by the willingness of persons to be examined in a roentgenographic survey if one were to take place.

Polls	Per cent answering "would be X-rayed"
Mishawaka	86
Silver Spring Poll 1	82
Silver Spring Poll 2	83

It would appear that the campaign to examine all adults in the United States is making headway, as reflected by the following:

Polls	Per cent who have been X-rayed
Gallup	42
Hartford	47
Denver	63
Mishawaka	57
Silver Spring Poll 1	64
Silver Spring Poll 2	89

The above responses probably reflect the extent to which opportunity has been afforded the various populations to have a chest roentgenogram.

COMMENT

The results of this study indicate that people's attitudes are, for the most part, constructive, and that the general level of knowledge is encouragingly high, expecially in regard to the contagious nature of tuberculosis and the fact that it can be cured. The great knowledge deficiencies concerned the inheritability of tuberculosis, the lack of symptoms in early tuberculosis, climate as a factor in the cure of tuberculosis, and the amount of tuberculosis in older age groups.

It was desired to find out, not only what misconceptions the people might have, but in what population groups the level of knowledge was lowest. The following generalities can be stated:

(1) Men and women have approximately the same degree of knowledge.

(2) Older people are less well informed than their younger neighbors.

(*3*) College people are better informed than high school people, who are, in turn, better informed than those with only an elementary school education.

(*4*) Persons with tuberculosis in their families know less than average on poll 1 but more than average on poll 2.

The improvement in people's knowledge during the roentgenographic survey was slight, only 2.3 per cent. However, greater improvement can be noted in some sections of the population and on some of the questions, *e.g.*, in age groups over 45, in persons with grammar school education or less, in persons with tuberculosis in their families, and most notably on questions having to do with roentgenographic examination. The fact that persons with tuberculosis in their families improved in knowledge regarding tuberculosis emphasizes the receptiveness of this group and points up the need for more intensive education.

A comparison was made of the results of the Silver Spring polls with those of similar polls conducted by the Gallup Poll and in the cities of Hartford, Connecticut; Denver, Colorado; and Mishawaka, Indiana. The most striking thing about such a comparison is the similarity of knowledge in these widely separated cities.

It would seem that during a roentgenographic survey, people's ideas about tuberculosis did not change radically, even though their ideas with regard to roentgenographic examinations did. It may be that, if knowledge has been increased in this one area, an important accomplishment has been made. Advertising specialists concentrate on one thing at a time, as an effort to cover a multitude of points accomplishes little. Education is a cumulative experience. To accomplish desirable objectives, tuberculosis education must be continuous.

It seems somewhat difficult to change preconceived ideas. It might even be said that an intermediate step to complete change is one of doubt. This concept would reemphasize the importance of the right idea in the first place and make the school of paramount importance in a community education plan.

Evaluation is an important part of any activity. The results reported above have furnished figures on the level of tuberculosis knowledge in Silver Spring on two different occasions, and have shown what can be expected in increased knowledge through an educational campaign such as accompanied the Montgomery County chest roentgenographic survey. Equally important, the facts are now available to furnish a sound foundation for future program planning.

SUMMARY

Two public opinion polls, one preceding and one following a county-wide chest roentgenographic survey, were conducted in Silver Spring, Maryland, in an attempt to find out what people learn about tuberculosis in the course of a roentgenographic survey. At the same time an attempt was made to obtain information of the people's knowledge and attitude concerning tuberculosis for use in the planning of future programs. Every step, the design of the questionnaire, the selection of the sample, the training of the volunteer interviewers, and the analysis of the responses, was carefully planned and executed in order that the results should be statistically accurate.

The improvement in people's knowledge during the roentgenographic survey was slight, only 2.3 per cent. Nevertheless improvement greater than this was noted in some sections of the population and in the responses to certain questions. Improvement was most evident in age groups over 45, in persons with grammar school education or less, in persons with tuberculosis in their families, and particularly in the answers to questions concerned with roentgenography.

SUMARIO

¿Qué Enseña al Público una Encuesta Radiológica?

Dos censos de la opinión pública, uno antes y otro después de una encuesta radiográfico-torácica en un distrito completo, fueron tomados en Silver Spring, Máryland, E. U. A., tratando de averiguar lo que la gente aprende acerca de la tuberclosis en el curso de una pesquisa radiográfica. Procuróse al mismo tiempo obtener información relativa a los conocimientos y actitud de la gente con respecto a la tuberculosis, con mira a emplear los datos al proyectar obras futuras. A fin de que los resultados fueran estadísticamente exactos, se estudiaron y ejecutaron con cuidado todas las medidas tomadas, el plan del cuestionario, la selección de la muestra, la preparación de los entrevistantes voluntarios y el análisis de las respuestas obtenidas.

El aumento de los conocimientos del público durante la pesquisa radiográfica fué leve: apenas de 2.3 por ciento, aunque se notó un aumento mayor que éste en algunos sectores de la población y en las respuestas a ciertas preguntas. El aumento fué más manifiesto en los grupos de más de 45 años de edad, en las personas con educación primaria o menos, en las que tenían tuberculosis en la familia, y particularmente en las respuestas a las preguntas referentes a roentgenografía.

ORAL PROTEIN HYDROLYSATE IN PULMONARY TUBERCULOSIS

A. A. KINNEAR AND E. N. KEEN[1]

(Received for publication May 13, 1948)

This paper records the results of feeding protein hydrolysate to colored adult males with tuberculosis. Thirty-eight patients were studied in all, 19 were "test" patients and 19 were "control" patients. To preserve random sampling, alternate patients were allotted as "test" patients.

PLAN OF INVESTIGATION

Complete clinical and radiological examinations revealed no evidence of other diseases. The urine of all patients was essentially normal except for one instance of transient albuminuria which later cleared. No evidence of amyloidosis was detected.

"Control" patients and "test" patients alike were submitted to the following procedures:

(1) The weight was recorded at 21 day intervals.
(2) The daily temperature was routinely recorded.
(3) The blood sedimentation rate was determined at 21 day intervals.
(4) A roentgenogram of the chest was taken at 21 day intervals.
(5) Ten ml. of blood were drawn from an arm vein with minimal constriction, also at 21 day intervals. Of this, 5 ml. were put into a tube with the correct amount of standard Wintrobe anticoagulant mixture. This portion was used to determine the packed cell volume, the hemoglobin concentration and hence the mean corpuscular hemoglobin concentration. The remaining 5 ml. of blood were allowed to clot and the serum separated. Part of this serum was fractionated and the remainder was used for the empiric liver function tests.

Each "test" patient was then given 50 Gm. of protein hydrolysate[2] per day over and above his ordinary hospital diet. This protein hydrolysate yielded 53 per cent protein on triplicate Macro-Kjeldahl determinations. At three weeks from the commencement of feeding and again at six weeks, blood was withdrawn as before and the investigations outlined above were repeated. The unpalatability of the preparation caused no difficulty.

Methods: The erythrocyte sedimentation rate was determined in Westergren tubes and the fall at the end of one hour was corrected for anemia by means of the packed cell volume.

The packed cell volume was determined in a standard Wintrobe (1) tube, after centrifuging for 30 minutes at 3,000 r.p.m.

Hemoglobin was determined by an alkaline hematin method. A Hilger photoelectric absorbiometer was calibrated with the inorganic solution of Gibson and Harrison (2), using a green filter at 530 μ. The resulting graph was used in reading hemoglobin values, estimations of which were done at least twice for each specimen.

[1] From the Union Health Department Laboratories, Cape Town, South Africa.

[2] Carnesco, a brand of protein hydrolysate was kindly made available through the good offices of Dr. B. A. Dormer, Chief Tuberculosis Officer for the Union, and the Natal Branch of the S. A. Red Cross. The formula supplied with the Carnesco A was: meat flour 40 per cent, dried skim milk 40 per cent, sugar 16 per cent, food yeast 4 per cent.

Total serum proteins were estimated after the method of Koch and McMeekin (3), using a Micro-Kjeldahl technique with subsequent nesslerization. Duplicate estimations were made and results differing by more than 0.2 Gm. were discarded.

Serum globulins were precipitated by the method of Howe (4), using 23 per cent sodium sulphate, and an aliquot of the albumin remaining in solution was subjected to a micro-Kjeldahl procedure with nesslerization. As above, duplicate estimations were made and results differing by more than 0.2 Gm. were discarded.

The thymol turbidity test was essentially that of Maclagan (7), using a thymol buffer of pH 7.8 and using the upper limit of normal as 4 Maclagan units.

The serum colloidal gold test (5) used was the modified Melagan technique (6), using a citrate sol. A buffer of pH 7.7 to 7.8 precluded the necessity for preliminary standardization of the constant particle gold sol.

The modified cephalin cholesterol flocculation test of Hanger (8, 9) was used, using both cephalin prepared in this laboratory and the combined cephalin cholesterol mixture made by Difco Co. No significant differences were noted in comparing the results of the two antigens. The serum for this test was separated without undue exposure to light, as has been emphasized by Neefe (10). Krautman (11) used an alkalized saline solution and found that nonspecific results were kept to a minimum. Alkalized saline (0.85 per cent) was used and positive and negative control sera were tested in each series of determinations.

RESULTS

Intensive study of the clinical records and the chest roentgenograms of the patients in the treated group revealed no measurable effects which could be attributed to the protein hydrolysate.

The results are presented in table 1. Under each heading the first line enumerates the results which fall within generally accepted normal limits. The next line or lines indicate degrees of abnormality as set out on the left. The results for both the "test" and "control" groups are presented in the three columns. The first column indicates the beginning of the survey, the second the middle (after three weeks), and the third, the position at the end of the six weeks survey period. It will be noted that some values are missing from the third column in the control section. This is due to the death of 2 of these patients and the departure of one other. Patients were allotted as far as possible alternately to the "control" and the "test" groups. The only bias entering into the selection was a temptation to choose the more cooperative type of patient for the test group. The results showed that the patients in the "control" group were, from the start, in a poorer nutritional state than those in the "test" group. Since, however, no significant change was recorded in the "test" group over the six weeks' period this fact becomes of less importance. The results have been pooled in order to study some aspects of the nutritional state of patients suffering from pulmonary tuberculosis.

Erythrocyte sedimentation rate: Of 58 patients at the beginning of the survey, 7

had erythrocyte sedimentation rates of 10 mm. or less. This proportion did not significantly alter during the six weeks, and in general, the rate neither altered nor fluctuated to any great extent in patients from either group. Getz, West, and their associates (12) found a positive statistical correlation between the sedimentation rate and the serum albumin level. An opportunity was taken to confirm these findings.

Over a total of 111 observations, a positive correlation of 0.60 was found. The fact that the amount of plasma albumin is one of the factors influencing the rate of clumping and sedimentation of red blood corpuscles was further illustrated by Gray and Mitchell (13), who showed that albumin added to blood with a rapid erythrocyte sedimentation rate slowed the rate down. These results, however, did not rule out the possibility that other protein fractions may have important effects on the sedimentation rate.

Packed cell volume: The proportion of patients (20 of 38) in whom the packed cell volume did not fall below 45 per cent must be compared with the figures for hemoglobin as given below. It would seem to indicate that the tuberculous state has rather less effect on the production of an adequate volume of red cells than it has on hemoglobin metabolism.

Hemoglobin: A hemoglobin concentration of 14 Gm. per 100 ml. was arbitrarily chosen as the lowest normal value. Twenty-four of the 38 patients failed to reach this level. This standard is probably less stringent than that set for packed cell volume and a higher figure would obviously have brought out the difference to a greater extent. Getz, West, and their associates (12) showed that the most constant deficiency finding in a series of adult males with tuberculosis was vitamin C deficiency. After that came hemoglobin deficiency and then plasma protein deficiency, especially in the albumin fraction of the serum protein. The present figures tend to confirm the hemoglobin findings although over one-third of the patients had more than 14 Gm. of hemoglobin per 100 ml. of blood.

Mean corpuscular hemoglobin concentration: As might be expected from the data already presented, the values for the mean corpuscular hemoglobin concentration showed striking reduction. The accepted normal lower limit of hemoglobin saturation is 32 per cent and only two estimations throughout the survey fell at or above this level. A poor hemoglobin concentration can thus be said to be the only consistent finding in all of the patients of this survey and is the point in hemopoiesis which the tuberculous toxemia appears to effect. The anemia produced is of the type classically supposed to result from disorders of iron metabolism resulting in poor hemoglobin synthesis. These patients had adequate iron in their diet and were not subject to repeated hemoptyses. Globin, a protein of the histone class, constitutes about 96 per cent of the hemoglobin molecule and it may be assumed that availability of protein is a necessity in the synthesis of hemoglobin. Orten and Orten (14) studied the formation of hemoglobin following the administration of certain amino acids to rats fed a diet low in protein. They concluded that no single amino acid was of paramount importance in hemoglobin synthesis, but rather that a combination of amino acids

in unknown proportions was necessary for the elaboration of hemoglobin. The results of the present study tend to show that protein available as the hydrolysate by itself had no effect in enhancing the hemoglobin levels.

In view of these facts, it was thought worth while to compare the serum protein levels with the values for the hemoglobin and packed cell volumes. No statistical correlation could be found between the total protein values and either of the other two measurements but, when albumin levels were compared, the following positive correlations were found. When serum albumin levels and hemoglobin content of the blood (105 observations) were compared, a positive correlation of 0.57 with a standard error of ±0.067 was found. The results for serum albumin levels and packed cell volume (105 observations) was 0.50 with a standard error of ±0.073.

Each of these figures is significant and the difference between them is also significant. These figures clearly show that there is a relationship between the factors bearing on the level of serum albumin and the hemoglobin level, at any rate in those patients studied. A similar and lesser relationship is shown between the serum albumin level and the formation of red blood cells. The precise nature of these relationships is not clear, but it is tempting to speculate that the answer may be found in the synthesis of globin, at any rate so far as the synthesis of hemoglobin is concerned.

Total serum protein: The great majority of estimations fell within the normal limits chosen (6 to 8 Gm. per 100 ml.). This observation is of little value when the figures after fractionation are examined. Here it is apparent that in many cases a fall in serum albumin is accompanied by a rise in serum globulin as may be expected if the level of serum globulin is in part a measure of gamma globulin concentration and probably antibody formation. In fact, Kunkel (15) suggests that nearly every increase in serum globulin in states of hyperglobulinemia is due to an increase in the gamma globulin fraction of the serum. The relation of antibodies to this fraction of the serum is well known and lends itself to interesting speculation on the significance of altered globulin ratios in infectious diseases which are not primarily hepatic.

Serum albumin: At the beginning of the survey, 13 of the 38 patients had albumin levels of less than 3.6 Gm. per 100 ml., which was arbitrarily chosen as the lower limit of normal. The relation of the serum albumin values to hemoglobin level, the packed cell volume, and the blood sedimentation rate has already been discussed. The relation to the empiric liver function tests will be discussed later.

Serum globulin: Twenty, or approximately half, of the patients showed elevated serum globulin levels. This was an expected finding (*vide supra*).

Liver function tests: Three empiric liver function tests were carried out on every specimen of serum collected in order to attempt an estimate of the functional state of the liver. Of the three tests, cephalin cholesterol flocculation, serum colloidal gold test and the thymol turbidity test, only the last named showed any deviation from normal. In this test half the patients gave a reading of 5 or over. It is well known, however, that under certain conditions, if the

ionic strength of a solution is reduced, the solubility of certain proteins is reduced and they are precipitated. This principle is used to advantage in both the thymol turbidity test and the buffer dilution test of Wolff. The fact that the thymol turbidity test is probably associated with an excess of beta globulin in the serum, and also that certain lipemic sera show a marked increase in the beta globulin fraction, adds a complicating factor to the interpretation of the thymol turbidity test taken by itself as an indication of liver dysfunction.

Although clinical evidence of hepatic insufficiency is rare in tuberculosis, the more sensitive tests show that functional derangement is not uncommon. A series of patients examined by Kruger and Gerber (16) and Hurst *et al.* (17) showed abnormalities in a proportion of patients, predominantly in the very advanced phases of pulmonary tuberculosis. It is interesting to note that Hurst and his colleagues failed to show changes in hepatic function with both the oral and intravenous hippuric acid test.

The present results with the flocculation tests do not seem to confirm the finding of liver derangement in pulmonary tuberculosis. Only a small number of tests were carried out, however, and it is a well-known fact that functional ability of the liver cannot be measured by any single functional test; a veritable battery of tests measuring different functions seems to be the only safe measure. Maclagan (20), using the flocculation tests, found one positive test in 18 patients suffering from pulmonary tuberculosis. It is interesting to note also that significant deviations from the normal were only consistently obtained with the serial bromsulfalein test. If some of these empiric tests owe their positivity to the excess of gamma globulin in the serum, and good antibody response is likewise associated with an increased gamma globulin fraction of the serum, the interpretation of the increased gamma globulin fractions cannot be construed as evidence of liver disease in infectious diseases.

COMMENT

The original purpose of the present study was to demonstrate the effect, if any, of adding protein hydrolysate to the diet of adult colored males with tuberculosis. Clinical data were studied and, in addition, roentgenographic examinations of the chest, erythrocyte sedimentation rates, and the blood analyses outlined above were carried out in order to measure any change in clinical or nutritional state. The reasoning behind the experiment was that some tuberculous patients found difficulty in digesting first-class protein, such as meat. The idea of giving such patients protein hydrolysate is not a new one. The liver is intimately concerned in protein metabolism and probably synthesizes proteins from amino acids supplied to it from the blood. Thus it seems reasonable to assume that the liver could continue this function if furnished with the necessary amino acids provided its own function was not grossly deranged by a disease process. For this reason empiric liver function tests were carried out on all sera and, as already stated, revealed very little evidence of liver dysfunction by themselves. Cox and Mueller (18) found that orally or intravenously administereed protein hydrolysate was equally effective in the regeneration of plasma albumin in hypoproteinic patients.

TABLE 1

Determinations of Various Components of the Blood during Protein Hydrolysate Therapy

	TEST GROUP NO. OF PATIENTS			CONTROL GROUP NO. OF PATIENTS		
	Beginning	Third Week	Sixth Week	Beginning	Third Week	Sixth Week
Erythrocyte Sedimentation Rate; mm. in one hour.						
0 to 10 mm.	5	7	6	2	—	—
11 to 50 mm.	10	9	9	5	8	9
more than 50 mm.	4	3	4	12	11	7
Packed Cell Volume, per cent						
45 and above	13	14	13	7	9	9
35 to 44	6	5	6	11	8	7
less than 35	—	—	—	1	2	—
Hemoglobin Gm. per 100 ml.						
14 Gm. and above.	9	7	9	5	3	5
12 to 13.9 Gm.	8	11	9	5	6	6
less than 12 Gm.	2	1	1	9	10	5
Mean Corpuscular Hemoglobin Concentration, per cent						
32 and above	1	1	—	—	—	—
29 to 31	14	10	15	11	12	8
less than 29	4	8	4	8	7	8
Total Serum Protein Gm. per 100 ml.						
6 to 8 Gm.	17	19	19	16	17	13
more than 8 Gm.	2	—	—	2	2	2
less than 6 Gm.	—	—	—	1	—	—
Serum Albumin, Gm. per 100 ml.						
3.6 to 5 Gm.	16	16	18	9	13	10
less than 3.6 Gm.	3	3	1	10	5	5
Serum Globulin, Gm. per 100 ml.						
1.3 to 3.2 Gm.	12	12	11	6	3	6
3.3 to 5 Gm.	7	7	8	13	15	9
Thymol Turbidity Test (units)						
0 to 4	10	9	9	9	7	7
5 to 7	9	10	9	9	10	5
8 to 10	—	—	1	1	2	3
Colloidal Gold Test (units)						
0 to 2	17	18	18	17	16	13
3 to 5	2	1	1	2	3	2
Cephalin Cholesterol Flocculation Test						
0 to +	17	19	18	17	17	13
2^+ to 4^+	2	0	1	2	2	3

Madden and Whipple (19) showed that amino acids as the sole source of nitrogen in standardized dogs could produce plasma proteins and the response compared favorably with the response of first class proteins in corresponding amounts.

The precise values in the observations of the present study can be seen in table 1 and are set out in table 2 in detail. Each patient was studied separately and the change in this nutritional state recorded. The mean values were calculated as well as the Standard Deviation for the variance of the changes. The standard error of the mean was calculated from the formula $\frac{S.\ D.}{n-1.}$ where "n" is the number of patients studied.

All the values indicate a very slight increase (including the serum globulin values), but the second column shows clearly that all these figures can be disregarded as being insignificant. The only possible exception is the mean increase in packed cell volume in the "control" group where the actual mean change was about twice its standard error. Such a result might occur by chance about once in twenty times. These data then fail entirely to show any measurable effect of the feeding of protein hydrolysates to these patients over a period of six weeks. The same conclusion was reached after intensive and careful study of the clinical and radiological records.

TABLE 2

Change in Values after Six Weeks

	MEAN CHANGE	STANDARD ERROR OF THE MEAN
Total Serum Protein		
Test group	+0.21 Gm.	±0.17 Gm.
Control group	+0.09 Gm.	±0.14 Gm.
Serum Albumin		
Test group	+0.09 Gm.	±0.13 Gm.
Control group	+0.05 Gm.	±0.12 Gm.
Serum Globulin		
Test group	+0.04 Gm.	±0.18 Gm.
Control group	+0.20 Gm.	±0.19 Gm.
Packed Cell Volume		
Test group	+0.6 per cent	±0.89 per cent
Control group	+1.25 per cent	±0.61 per cent
Hemoglobin		
Test group	+0.02 Gm.	±0.31 Gm.
Control group	+0.08 Gm.	±0.25 Gm.

SUMMARY

Thirty-eight patients with pulmonary tuberculosis were divided into two equal groups. One group received an additional dietary supplement of 50 Gm. of protein hydrolysate per day.

The results of this treatment were gauged by clinical, roentgenographic, and biochemical methods.

During the six weeks period of observation no beneficial effects of statistical significance were noted which could be attributed to the increased protein in the diet.

SUMARIO

Hidrolisato de Proteína por Vía Oral en la Tuberculosis Pulmonar

Treinta y ocho tuberculosos pulmonares fueron divididos en dos grupos iguales, recibiendo un grupo, como complemento dietético, 50 Gm. diarios de hidrolisato de proteína.

El resultado de este tratamiento fué justipreciado clínica, radiográfica y bioquímicamente.

Durante el período de observación de seis semanas no se observaron efectos beneficiosos de cuantía estadística que pudieran imputarse al aumento deproteína en la alimentación.

Acknowledgment

We wish to thank Prof. R. Turner for permission to publish this paper. We are indebted to Dr. J. F. Wicht, Medical Superintendent of Hospitals, for advice and interest. To Dr. F. Ames we are grateful for excellent supervision of the patients and collection of the blood samples.

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SUPPLEMENTARY ORAL PROTEIN HYDROLYSATE THERAPY IN TUBERCULOSIS[1]

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INTRODUCTION

The importance of adequate protein nutrition in a number of medical and surgical conditions has been frequently stressed in recent years. Protein deficiency may be due to an insufficient intake, poor absorption or assimilation, excessive protein destruction or loss. In infection, the degree and duration of destruction of protein depends on the severity and chronicity of the disease. The attempt to utilize protein for repair of damaged tissue takes precedence over and may occur at the expense of the catabolism of normal tissue. Consequently, it is probable that the ordinary hospital diet may not at times be adequate to supply this combined demand for protein. Also, a higher than usual caloric intake may be necessary because, if fats and carbohydrates are not present in sufficient amount, protein will be burned for heat. The quality of protein as well as its quantity is important too. Animal protein is generally preferable to vegetable protein. A deficiency of protein may be due to an insufficiency of protein per se or of specific amino acids. Of the twenty-one amino acids, eight are considered as essential in man for the formation of protein and other nitrogenous compounds. These essential amino acids cannot be synthesized by the body but must be supplied in the food.

An experiment was undertaken in a group of patients with far advanced tuberculosis to supplement the regular hospital dietary with the oral use of amino acid mixtures available in protein hydrolysates. An attempt was made to correlate the value of this nutritional regimen with clinical observations and laboratory determinations.

MATERIALS AND METHODS

Selection of patients: Patients were selected who fulfilled the following criteria: (*1*) residence in hospital for at least six months prior to the supplementary use of protein hydrolysates; (*2*) evidence of unfavorable clinical and/or roentgenological course with sputum positive for *M. tuberculosis*, weight loss, and fever. Attempts at collapse therapy had been unsuccessful in these patients or they were considered unsuitable for such therapy when the study was begun.

During the preliminary observation period, the patients received a general diet which in this hospital ranges from 2,500 to 2,700 calories with an average protein allowance of 70 to 90 Gm. In addition, the patients received between meal nourishment of three glasses of milk daily or an approximate equivalent substitute depending on the patients' preference, and also a supplementary intake of vitamins.

Clinical and laboratory studies: A physical examination was done at the onset and repeated as indicated. Sputum examinations were ordered as necessary. Roentgenograms of the chest were taken before therapy and at approximately three month intervals. The

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maximum and standard normal (for corresponding age, sex, height) weight records were noted on admission and at weekly intervals thereafter. The additional laboratory determinations which were made prior to and at one to three month intervals during therapy included: (1) hematocrit (Haden method); (2) erythrocyte sedimentation rate (Cutler method); (3) concentration of serum protein and the albumin globulin ratio (Reiner method); (4) blood volume with T-1824 dye (Gibson and Evans method (1), modified by Nobile *et al.*, (2)); and (5) the plasma volume and the total circulating protein which were calculated from the above determinations.

Protein hydrolysate products: Three preparations were employed: (*1*) Protolysate, a dry enzymic digest of casein containing amino acids and polypeptides; (*2*) Edamin, a tryptic hydrolysate of milk protein which consisted primarily of amino acids and peptides derived from lactalbumin; (*3*) Granules-Protein-Mineral Mixture, a protein of liver origin subjected to partial digestion with added minerals. The respective total nitrogen percentage content of each preparation was 12.0, 12.0, 7.0.

Method of hyperalimentation therapy: The daily hydrolysate intake for each patient was calculated according to the following formula. The total daily nitrogen requirement based on the patient's observed weight was computed on the arbitrary basis of 0.6 Gm. per Kg. and the known nitrogen content of the proprietary product used. The total caloric intake was estimated using 40 calories per Kg. as the standard. Since one gram of protein hydrolysate yields 4 calories, the protein caloric intake was available. To make up the deficit in the total calories, Dextri-Maltose #2 (without salt) was employed. It is palatable and acts as a protein sparer or protector of body protein, relieving tissue protein of the necessity of furnishing energy. For example, a patient weighing 60 Kg. will need 36 Gm. of nitrogen daily. Assuming that protolysate is used, 12 Gm. of nitrogen are furnished by 100 Gm. of this product, so that a total of 300 Gm. is necessary. This furnishes 1,200 calories. The total caloric requirement of this 60 Kg. individual is 2,400. Therefore, the caloric deficit (2,400 minus 1,200) is 1,200 which is furnished by the 300 Gm. of Dextri-Maltose #2.

In the preparation of the mixture, the latter was first dissolved in one or one and one-half liters of boiled water to which the specific protein hydrolysate product was then added. The mixture was prepared fresh daily and kept in a refrigerator. Four or five divided oral feedings were given before or after meals according to the patients' preference. In addition, each patient received a diet of about 2,500 calories, containing 75 Gm. of protein.

RESULTS

A total of 9 patients with far advanced tuberculosis were placed on the hyperalimentation diet. Three died from their basic disease very soon after therapy was begun and should properly be excluded for any fair evaluation of this mode of therapy. Six patients were maintained on the hyperalimentation regimen over a period ranging from approximately six to eleven months. Four of the surviving patients were then placed on the routine hospital diet and final observations were recorded six months later. A control period was therefore available before and after hyperalimentation was instituted. A summary of the pertinent observations in each of the 6 cases is presented.

CASE REPORTS

Case I: F. L., a 36 year old white male (figure 1) was known to have had pulmonary tuberculosis since 1930. In 1933 he had a two stage thoracoplasty which was ineffective. His last hospital admission occurred in May, 1943 because of increased pulmonary symptoms and a loss of 20 lbs. in five months. The patient's weight on admission was 126.5 lbs. and on a regimen of bed rest (further surgical therapy was contraindicated) and the ordinary

hospital diet, his weight had declined to 102 lbs. on July 1, 1946. The temperature ranged up to 100°F. On September 16, amino acid therapy (Edamin and Dextri-Maltose 240 Gm. each) supplementing a regular diet containing 75 Gm. of protein was initiated. The weight was now 104 lbs., the sputum was positive for tubercle bacilli and a chest roentgenogram,

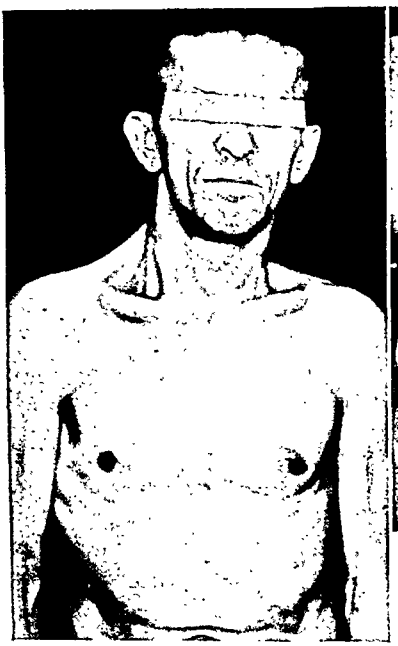


FIG. 1. (Left) Case I (F. L.). September, 1946
FIG. 2. (Right) Case I. September 10, 1946

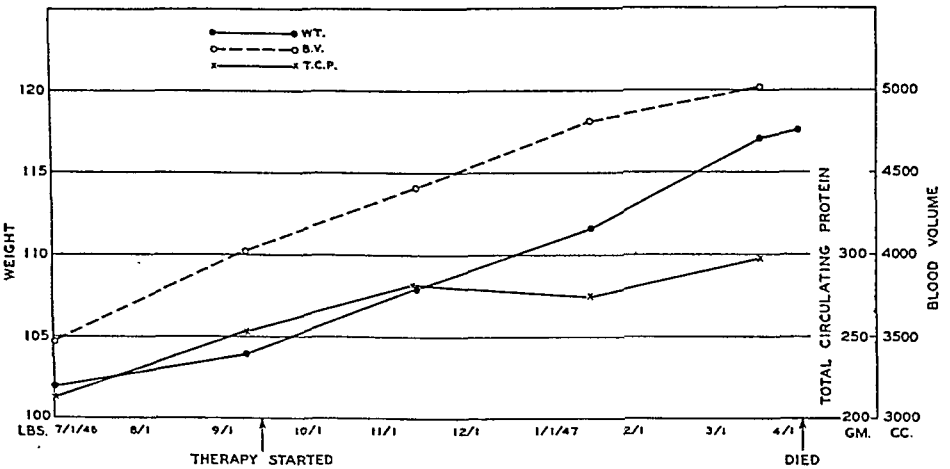


FIG. 3. Case I. Relation of blood volume, weight and total circulating protein

September 10, 1946, (figure 2) revealed evidence of a right sided thoracoplasty, bilateral productive disease with cavitation, a recent bronchopneumonic spread on the left, and adhesive pleurisy. The patient exhibited no significant untoward gastro-intestinal symptoms during the period of dietary therapy. He made no serious objection to the taste of the amino acid preparation.

The patient had recurrent small hemoptyses from October, 1946 through March, 1947. A roentgenogram of the chest in November, 1946 showed some resolution of the exudative process on the left. No significant change was observed in a film taken in February, 1947. On April 3, 1947, the patient suddenly developed dyspnea, rapid pulse, and died. A post-

TABLE 1

Summary of Data, Case I (F. L.)

DIET	DATE	WEIGHT	E.S.R.	HEMATOCRIT	SERUM PROTEINS	PLASMA VOLUME	BLOOD VOLUME	TOTAL CIRCULATING PROTEINS
		lbs.			*Gm.*	*cc.*	*cc.*	*Gm.*
	5/11/43 (Admitted)	126.5	—	—	—	—	—	—
Regular	7/ 1/46	102	26	24	T.P.—7.1 A.—4.5 G.—2.6 A/G—1.7:1	2,999	3,496	213
	9/10/46	104	26	22	T.P.—8.7 A.—4.9 G.—3.8 A/G—1.3:1	3,125	4,006	253
Regular plus oral amino acid therapy Edamin—240 Gm. D.M.—240 Gm. Started 9/16/46	11/14/46	108	28	23	T.P.—8.1 A.—4.0 G.—4.1 A/G—0.97:1	3,423	4,419	280
	1/16/47	111.5	24	24	T.P.—7.5 A.—4.5 G.—3.0 A/G—1.5:1	3,660	4,810	275
	3/18/47	117	26	24	T.P.—7.8 A.—4.8 G.—3.0 A/G—1.6:1	3,813	5,017	297
	4/ 2/47	117.5 Last recorded weight	—	—	—	—	—	—
	4/ 3/47 (died)							

mortem film revealed a considerable density on the left indicative of probable atelectasis of the left lung.

Prior to death, the patient showed gratifying constitutional improvement for which the new dietary regimen could receive much or all of the credit. His last recorded weight on

April 2, 1947 was 117.5 lbs. which represented a gain of 13.5 lbs. during the more than six plus months of supplemental amino acid therapy. The increase in the weight was paralleled by an increase in blood volume and a slightly less marked increase in the total circulating protein. The hematocrit determinations were consistently subnormal and significantly

TABLE 2

Summary of Data, Case II (R. T.)

DIET	DATE	WEIGHT	E S.R.	HEMATOCRIT	SERUM PROTEINS	PLASMA VOLUME	BLOOD VOLUME	TOTAL CIRCULATING PROTEINS
		lbs.			*Gm.*	*cc.*	*cc.*	*Gm.*
	1/ 7/46 (Admitted)	123	—	—	—	—	—	—
	Apr./46 (max.)	143	—	—	—	—	—	—
Regular	7/ 1/46	129.5	28	22	T.P.—7.5 A.—4.7 G.—2.8 A/G.—1.6:1	3,390	4,346	254
	9/10/46	121	26	21	T.P.—7.9 A.—4.9 G.—3.0 A/G.—1.6:1	3,129	3,960	247
Regular plus oral amino acid therapy Edamin—275 Gm. D.M.—275 Gm. Started 9/16/46 Protein Granules (200 Gm.) Substituted 12/20/46	11/14/46	116	24	21	T.P.—7.5 A.—4.1 G.—3.4 A/G.—1.2:1	3,662	4,637	274
	1/16/47	108	29	20	T.P.—7.2 A.—3.6 G.—3.6 A/G.—1:1	3,280	4,100	236
	2/ 5/47	105 Last weight record	—	—	—	—	—	—
	3/ 8/47 (died)	—	—	—	—	—	—	—

unchanged. The total serum protein concentrations remained within normal limits although the albumin-globulin ratio was depressed (table 1, figure 3).

Case II: R. T., a white male, age 43 (figure 4), was admitted January 7, 1946 with bilateral far advanced infiltrative and cavernous tuberculosis with sputum positive for tubercle bacilli. On bed rest his weight increased from 123 lbs. on admission to a maximum of 143 lbs. in April, 1946. A chest roentgenogram in September, 1946 (figure 5) showed dis-

seminated bilateral disease with multiple cavities and diaphragmatic pleurisy. The sputum remained positive for *M. tuberculosis* and the average daily febrile peak was 101°F. On September 16, 1946, a 75 Gm. protein diet was supplemented with Edamin and Dextri-Maltose, 275 Gm. each. The patient's weight was then 121 lbs. He experienced frequent

FIG. 4. (Left) Case II (R. T.). September, 1946
FIG. 5. (Right) Case II. September, 1946

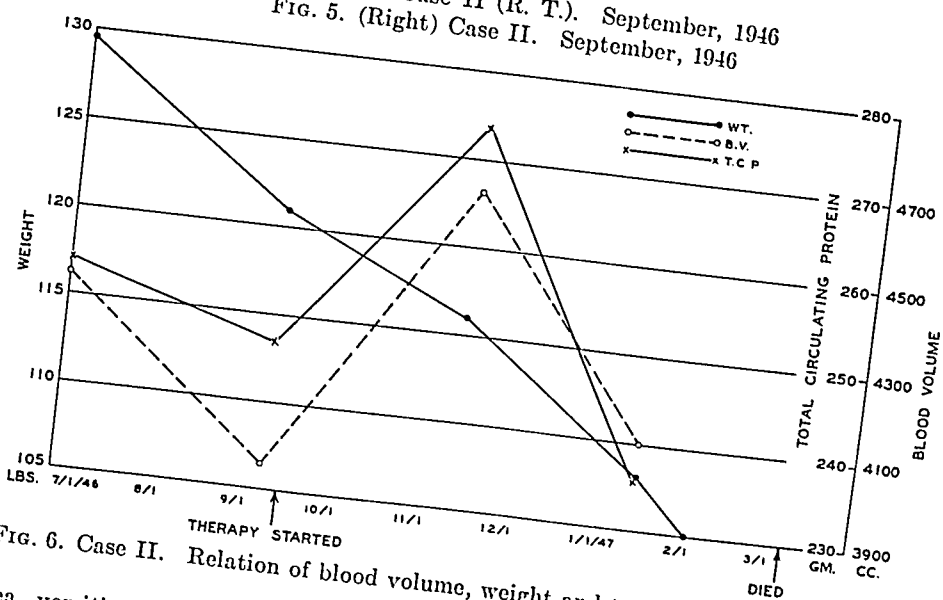


FIG. 6. Case II. Relation of blood volume, weight and total circulating protein

nausea, vomiting, abdominal cramps and occasional diarrhea. On December 20, 1946 Protein Granules (200 Gm.) was substituted with some abatement of symptoms. However, amino acid therapy had to be reduced in quantity or discontinued for short intervals. The patient frequently could not ingest all of his regular diet either. He gradually became

weaker, lost weight and his disease progressed, as determined on subsequent roentgenograms. The patient died March 9, 1947 after a therapy period of approximately six months.

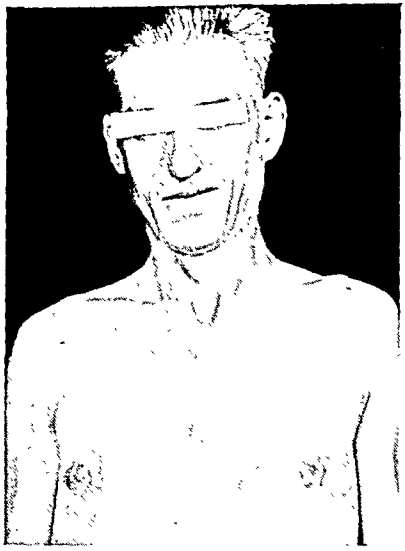
TABLE 3

Summary of Data, Case III (P. D.)

DIET	DATE	WT.	E.S.R.	HEMATOCRIT	SERUM PROTEINS	PLASMA VOLUME	BLOOD VOLUME	TOTAL CIRCULATING PROTEINS	LIVER FUNCTION TESTS
		lbs.			*gm.*	*cc.*	*cc.*	*gm.*	
	1/25/43 (Admitted)	Not recorded	—	—	—	—	—	—	
	Aug./43	136	—	—	—	—	—	—	
Regular	9/24/46	142.5	25	24	T.P. —7.7 A —4.5 G —3.2 A/G.—1.3:1	3,526	4,640	271	
	10/24/46	138.5	24	24	T.P. —7.5 A —5.3 G —2.2 A/G.—2.4:1	3,231	4,251	245	
	1/23/47	136.5	23	24	T.P. —7.8 A —5.4 G —2.4 A/G.—2.2:1	2,816	3,657	219	Bromsulfalein dye test 1 /30/47—6.5 per cent retention 12/18/47—Negative
Regular plus oral amino acid therapy Protolysate—310 Gm. D.M.—310 Gm. Started 10/28/46	4/23/47	145.5	22	22	T.P. —7.4 A —4.9 G —2.5 A/G.—1.9:1	3,521	4,512	260	
	7/ 8/47	148	25	21	T.P. —8.2 A —4.6 G —3.6 A/G.—1.2:1	3,817	4,831	312	Cephalin Flocculation 1/30/47—Negative 12/18/47—Negative
	9/16/47	152.5	23	24	T.P. —8.2 A —5.4 G —2.8 A/G.—1.9:1	4,453	5,859	345	
Regular from 9/17/47 to 3/10/48	12 16/47	151	23	25	T.P. —6.8 A —4.4 G —2.4 A/G.—1.8:1	3,650	4,866	248	
	3/10/48	140.5	23	22	T.P. —6.1 A —3.8 G —2.3 A/G.—1.6:1	3,497	4,483	214	

The dietary program was definitely unsatisfactory but his prognosis was hopeless from the beginning. There was poor correlation between the blood volume and weight records. The hematocrit was low, the serum protein concentration normal, but the albumin-globulin ratio was subnormal (table 2, figure 6).

Case III: P. D., a 43 year old white male (figure 7) was admitted February 25, 1943 with a history of pulmonary tuberculosis since January, 1942. On admission, the patient had hemoptyses, bilateral productive and cavernous disease, and sputum positive for *M. tuber-*



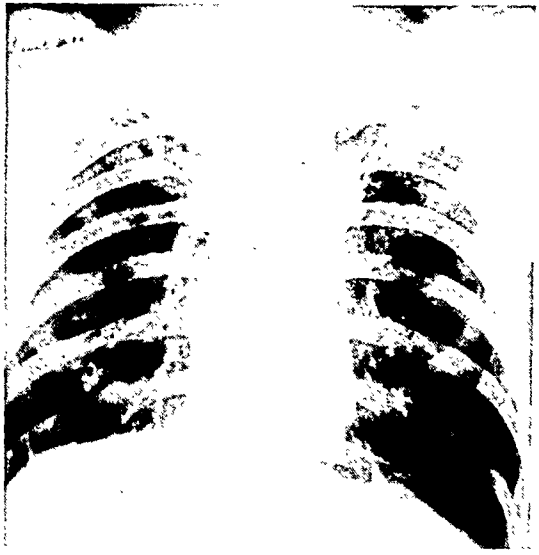


FIG. 7. (Left) Case III (P. D.). October, 1946
FIG. 8. (Right) Case III. October 4, 1946

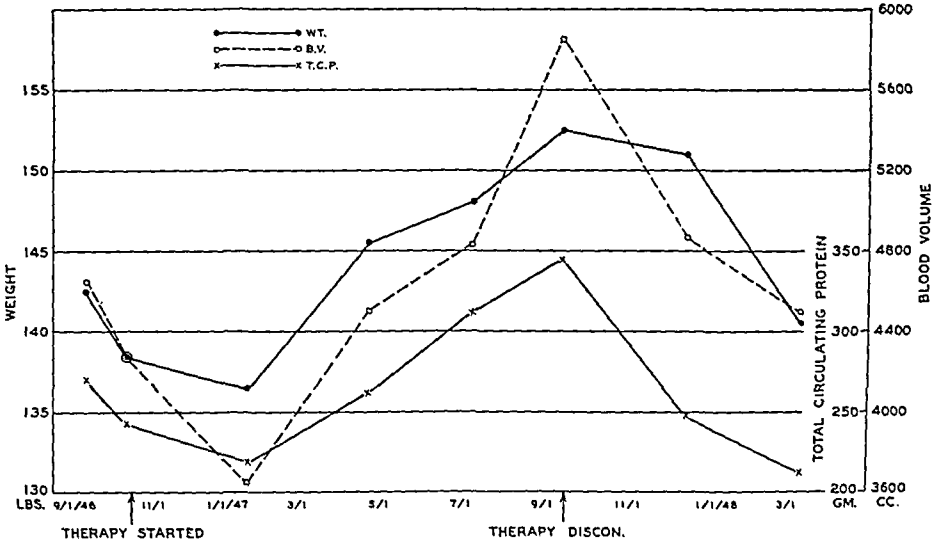


FIG. 9. Case III. Relation of blood volume, weight and total circulating protein

culosis. His standard normal weight was 160 lbs., the maximum 195 and the entrance weight was not recorded because of the pulmonary bleeding. In August, 1943 his weight was 136 lbs. and during the next three years he gained up to 142.5 lbs. (September 24, 1946), although his weight dropped to 138.5 lbs. one month later. A roentgenogram of the chest

(October 10, 1946) (figure 8) revealed that considerable clearing had occurred compared to the admission findings but residual bilateral emphysema with productive infiltration and multiple small cavities persisted in the upper lung fields. The sputum was positive for *M. tuberculosis* and he had the additional complications of amyloidosis (albuminuria, enlarged liver, 100 per cent retention of congo red in tissue) and hoarseness without dysphagia.

On October 28, 1946, the hyperalimentation regimen was begun consisting of the 75 Gm. protein diet plus Protolysate (310 Gm.) and an equal amount of Dextri-Maltose. During the first two months, the patient vomited about once every other day and had diarrhea for one day. His weight declined to 136.5 lbs. in January, 1947, but thereafter showed a steady increase to 152.5 lbs. on September 16, 1947, when the dietary therapy was discontinued after a period of approximately eleven months. In other words, his 14 lb. net gain in weight (from 138.5 to 152.5) in this latter interval was greater than had occurred in the three year previous period while on the ordinary hospital diet. No very significant change in the pulmonary process was noted on serial roentgenograms. The sputum remained positive for *M. tuberculosis*, albuminuria diminished, hoarseness persisted and another congo red test (October 3, 1947) showed no change. During supplemental amino acid therapy, there was a concomitant and significant increase in the plasma and blood volumes and the circulating protein; the serum protein and the albumin-globulin ratio remained essentially normal (table 3, figure 9). An abnormal value for the bromsulfalein test, which was noted about three months after treatment was started, became normal on completion of the dietary therapy.

The patient reverted to the original regular diet alone and observations were made in this control period during the six month interval between September, 1947 and March, 1948. He maintained almost the same weight for about three months and then a decline occurred to 140.5 lbs. The plasma and blood volumes and the circulating blood protein also diminished and the albuminuria increased (qualitative determination). Fluoroscopic and roentgenographic examinations of the chest in this period revealed no change.

The evidence strongly indicates that the addition and subsequent subtraction of the protein hydrolysate from the regular diet in this patient was directly related to the rise and fall in weight.

Case IV: J. M., a 41 year old white male (figure 10), entered the hospital April 10, 1941 with bilateral pulmonary tuberculosis and a large cavity in the left upper lobe. His weight was 110 lbs. and the highest recorded weight was 112 lbs. on May 28, 1946. A two stage thoracoplasty (left) was done September, 1946. Further operation was contraindicated because of poor respiratory reserve and a residual cavity and sputum positive for *M. tuberculosis* persisted (figure 11). Following thoracoplasty, the weight declined and on December 20, 1946 it was 99 lbs. at which time he was started on the diet containing 75 Gm. of protein and supplemented by Protolysate and Dextri-Maltose, 225 Gm. each. The patient's objection to the taste of the product was overcome by the addition of 4 cc. of peppermint water to the liter of mixture. He vomited once on the first day but at no time did he have abdominal cramps or diarrhea. Hyperalimentation therapy was stopped in September, 1947 after about nine months. At this time his weight was 107.5 lbs. which represented a gain of 8.5 lbs. The plasma and blood volumes and the circulating protein determinations fluctuated and showed no consistent correlation with the weight records (table 4, figure 12). Two liver function tests were normal.

During the second control period of six months with resumption of the regular hospital diet, the patient exceeded slightly the last recorded weight while he received Protolysate (107.5 to 109.5 lbs.). Serial roentgenograms showed some diminution in size of the residual cavity (left) both during amino acid therapy and subsequently, but the improvement was attributed to the bed rest regimen.

In summary, this patient tolerated well the hyperalimentation diet which was begun approximately three months after the thoracoplasty operation because of

TABLE 4

Summary of Data, Case IV (J. M.)

DIET	DATE	WT.	E.S.R.	HEMATOCRIT	SERUM PROTEINS	PLASMA VOLUME	BLOOD VOLUME	TOTAL CIRCULATING PROTEINS	LIVER FUNCTION TESTS
		lbs.			*gm.*	*cc.*	*cc.*	*gm.*	
	Apr./41	110 (admitted)	—	—	—	—	—	—	
Regular	11/19/46	97	26	22	T.P. —7.7 A —5.7 G —2.0 A/G.—2.8:1	2,077	2,663	159	
	12/19/46	99	21	26	T.P. —6.6 A —4.3 G —2.3 A/G.—1.8:1	1,982	2,678	130	
Regular plus oral amino acid therapy Protolysate—225 Gm. D.M.—225 Gm. Started 12/20/46	2/19/47	100.5	19	24	T.P. —6.5 A —4.0 G —2.5 A/G.—1.6:1	2,412	3,173	156	Bromsulfalein dye test 1/30/47—1.0 per cent retention 12/23/47—0.5 per cent retention
	4/24/47	101	11	20	T.P. —7.9 A —4.9 G —3.0 A/G.—2.6:1	1,832	2,410	144	
	7/ 8/47	102.5	20	22	T.P. —8.3 A —5.7 G —2.6 A/G.—2.2:1	2,040	2,615	217	Cephalin Flocculation 1/30/47—Negative 12/23/47—Negative
	9/14/47	107.5	19	23.5	T.P. —8.2 A —5.0 G —3.2 A/G.—1.5:1	1,723	2,252	141	
Regular (from 9/15/47 to 3/10/48)	12/16/47	109.5	15	27	T.P. —7.3 A —4.7 G —2.6 A/G.—1.8:1	2,139	2,930	156	
	3/10/48	109.5	16	23	T.P. —6.3 A —3.9 G —2.4 A/G.—1.2:1	2,366	3,072	149	

his slow constitutional recovery. An increase in weight followed and this tendency was maintained after amino acid therapy was stopped.

Case V: I. F., a 35 year old white female (figure 13) with a history of tuberculosis since 1937

and an unsuccessful period of bilateral artificial pneumothorax therapy, entered the hospital February 18, 1946. The sputum was positive for *M. tuberculosis* and the admission

FIG. 10. (Left) Case IV (J. M.). December, 1946
FIG. 11. (Right) Case IV. November 4, 1946

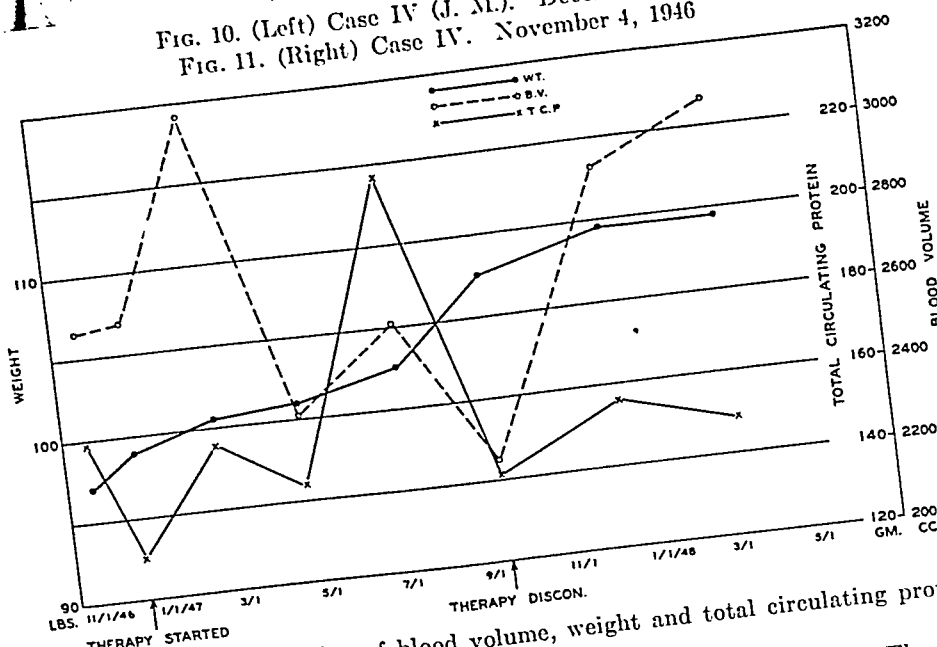


FIG. 12. Case IV. Relation of blood volume, weight and total circulating protein

film of the chest revealed bilateral disseminated infiltration with cavitation. The patient had an intermittent low grade fever. On bed rest, she gained from 101 lbs. on admission to a maximum of 116 lbs. in August, 1946, but the weight then dropped gradually to 105 lbs. in March, 1947. A chest roentgenogram (figure 14) showed in comparison to the admission

status slight resolution of the infiltrate in some areas but a definite increase in size of cavitation.

Hyperalimentation therapy was started March 21, 1947 with the addition to the usual diet of 240 Gm. each of Edamin and Dextri-Maltose. Occasional nausea and vomiting

TABLE 5

Summary of Data, Case V (I. F.)

DIET	DATE	WT.	E.S.R.	HEMATOCRIT	SERUM PROTEINS	PLASMA VOLUME	BLOOD VOLUME	TOTAL CIRCULATING PROTEINS	LIVER FUNCTION TESTS
		lbs.			*gm.*	*cc.*	*cc.*	*gm.*	
Regular	2/18/46	101 (admitted)	—	—	—	—	—	—	
	Aug./46	116 (max.)	—	—	—	—	—	—	
	3/20/47	105	25	21.5	T.P. —5.8 A —3.2 G —2.6 A/G.—1.2:1	2,861	3,621	165	
Regular plus amino acid therapy Edamin—240 Gm. D.M.—240 Gm. Started 3/21/47 Protein granules —350 Gm. D.M.—350 Gm. Started 5/12/47	4/24/47	104	27	20	T.P. —7.4 A —4.6 G —2.8 A/G.—1.3:1	2,355	2,945	174	Bromsulfalein dye test 3/23/47—8.5 per cent retention 12/29/47—Negative
	5/22/47	101.5	27	19	T.P. —8.1 A —4.5 G —3.6 A/G.—1.3:1	2,233	2:757	180	
	7/24/47	98	26	19.5	T.P. —8.4 A —5.7 G —2.7 A/G.—2.1	2,438	2,931	204	Cephalin Flocculation 3/23/47 + 12-29-47—Negative
	9/ 9/47	97	19	20	T.P. —8.1 A —4.7 G —3.4 A/G.—1.3:1	2,238	2,797	181	
Regular (from 9/10/47 to 3/12/48)	12/18/47	94.5	24	24	T.P. —7.8 A —4.6 G —3.2 A/G.—1.4:1	2,046	2,692	159	
	3/12/48	90	22	18	T.P. —7.0 A —4.1 G —2.9 A/G.—1.4:1	2,347	2,862	164	

occurred and, because the mixture became distasteful, 350 Gm. of Protein Granules were substituted on May 12, 1947. This product was more palatable but had to be discontinued on occasions. After approximately six months, the dietary regimen was stopped in September 1947. Progression of the pulmonary disease on the right side was noted and she lost 7 lbs. (from 104 to 97) during therapy. Further loss of weight occurred in the six months post-therapy observation period, 90 lbs. on March 12, 1948, although the roentgenogram of the chest showed slight regression of disease.

The blood volume and the circulating protein determinations for the most part did not parallel the weight records (table 5, figure 15). The result of the bromsulfalein test was definitely abnormal at the onset of treatment and later reverted to normal.

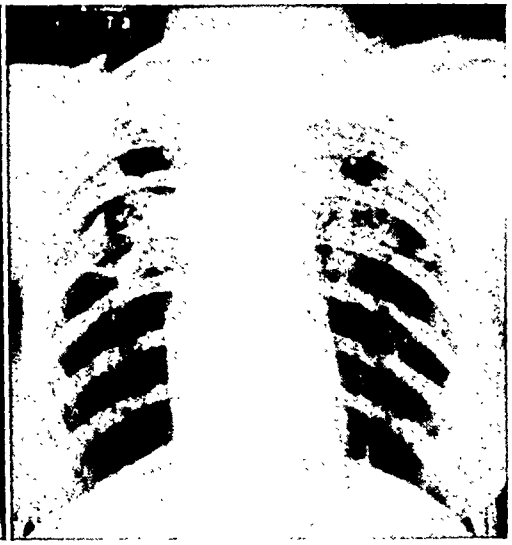


FIG. 13. (Left) Case V (I. F.). March, 1947
FIG. 14. (Right) Case V. March 3, 1947

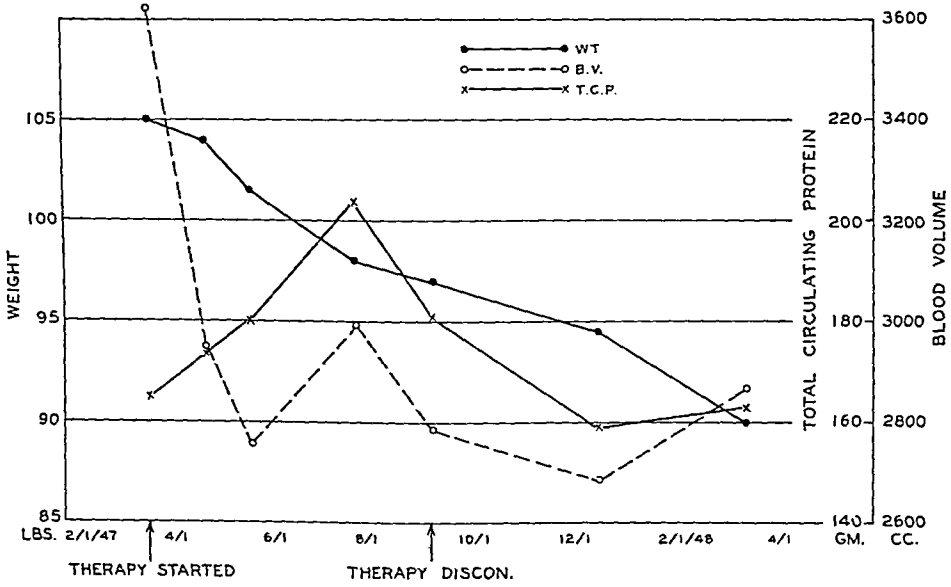


FIG. 15. Case V. Relation of blood volume, weight and total circulating protein

In summary, the results with hyperalimentation in this patient were poor both from the point of view of tolerance to it and from other objective findings. The unfavorable constitutional trend which was present before and during treatment also continued after its cessation.

Case VI: J. D., a 22 year old white male (figure 16) was readmitted November 18, 1946. The patient was febrile, in poor general condition, the sputum was positive for *M. tuberculosis* and a chest roentgenogram revealed a large upper lobe cavity (left) with bronchopneumonic disease and minimal infiltration (right). Artificial pneumothorax was induced but later discontinued. His weight on admission was 110 lbs., reached a peak of 115 lbs. in January,

TABLE 6

Summary of Data, Case VI (J. D.)

DIET	DATE	WT.	E.S.R.	HEMATOCRIT	SERUM PROTEINS	PLASMA VOLUME	BLOOD VOLUME	TOTAL CIRCULATING PROTEINS	LIVER FUNCTION TESTS
		lbs.			*gm.*	*cc.*	*cc.*	*gm.*	
Regular	11/18/46	110 (admitted)	—	—	—	—	—	—	
	1/13/47	115 (max.)	—	—	—	—	—	—	
	3/20/47	103	26	23	T.P. —7.7 A —4.0 G —3.7 A/G.—1.09:1	3,264	4,239	251	Bromsulfalein dye test 3/30/47—13 per cent Retention 12/29/47—1 per cent Retention
Regular plus Protein Granules —405 Gm. DM—405 Gm. Started 3/21/47	4/23/47	102	25	19	T.P. —7.2 A —5.2 G —2.0 A/G.—2.6:1	2,323	2,868	168	
	5/22/47	100	26	18.5	T.P. —7.4 A —5.3 G —2.1 A/G.—2.5:1	2,500	3,205	185	Cephalin Flocculation 3/30/47 ++ 12/29/47 0
	7/24/47	98	24	22	T.P. —8.0 A —5.4 G —2.6 A/G.—2.1	2,324	2,980	186	
	9/ 9/47	97	19	24	T.P. —7.8 A —4.9 G —2.9 A/G.—1.6:1	2,256	2,968	176	
Regular (9/10/47–3/12/48)	12/14/47	93	20	22.5	T.P. —6.5 A —4.4 G —2.1 A/G.—2:1	2,042	2,634	132	
	3/12/48	108.5	23	22	T.P. —7.4 A —5.3 G —2.1 A/G.—2.4:1	2,436	3,123	180	

1947 and then declined to 103 lbs. on March 21, when the hyperalimentation diet was initiated. The patient received 450 Gm. of the Protein-Granules-Mixture plus an equivalent amount of Dextri-Maltose and the associated regular diet. A roentgenogram of April 15, 1947 (figure 17) was used to visualize the status of the re-expanded left lung with its large cavity and fluid level. Occasional low grade fever was present.

It was hoped that the new diet might contribute toward sufficient improvement so that

surgical collapse therapy could be undertaken. Because a chest film in May, 1947 revealed more disease in the right lung, streptomycin therapy was started, and continued for six months. The patient lost 5 lbs. but roentgenologic improvement of the lesion occurred

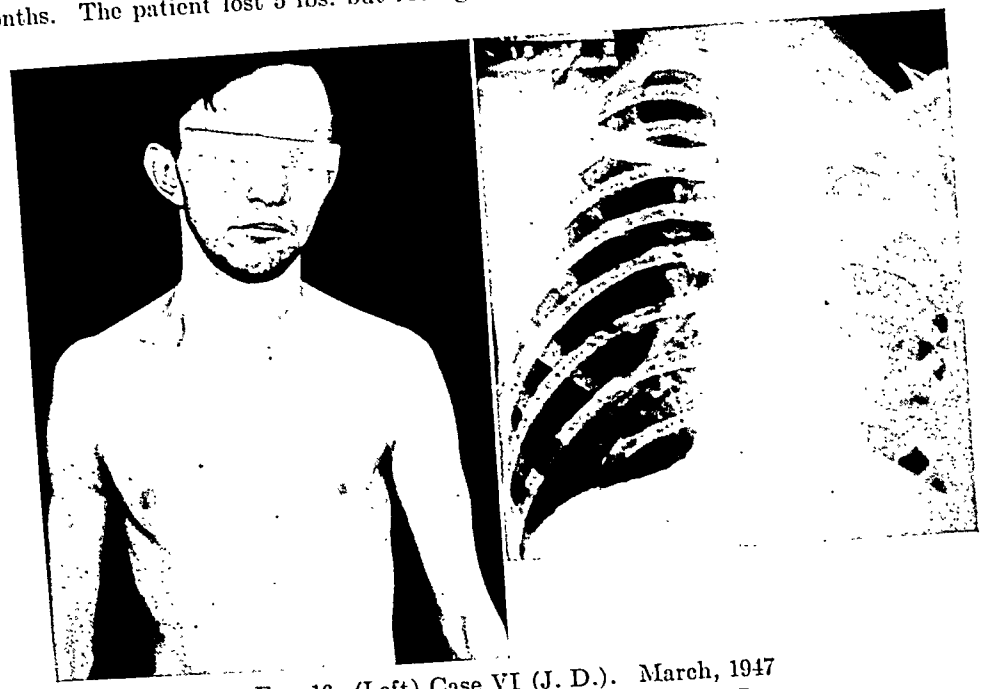


FIG. 16. (Left) Case VI (J. D.). March, 1947
FIG. 17. (Right) Case VI. April 15, 1947

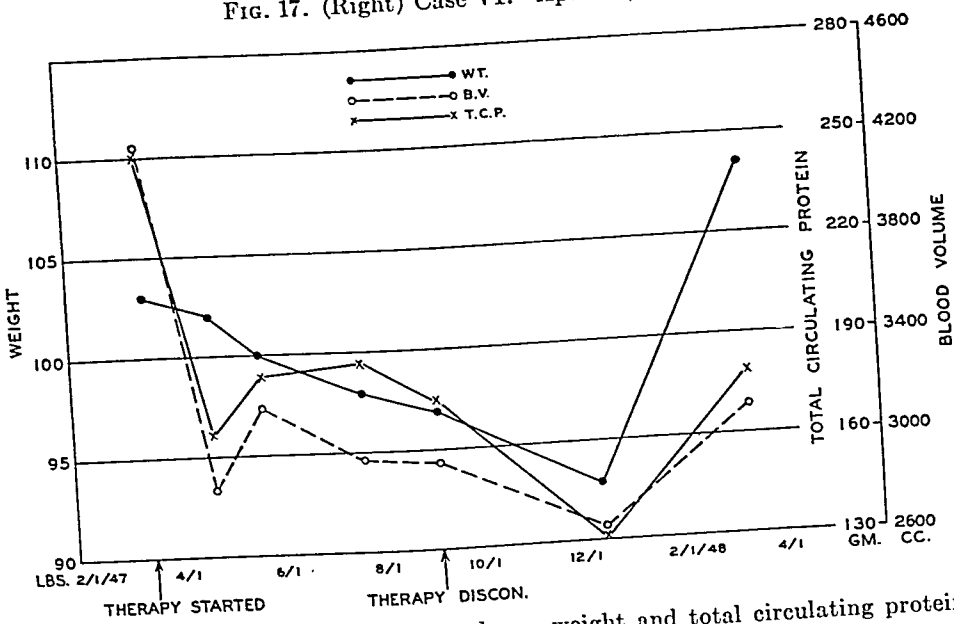


FIG. 18. Case VI. Relation of blood volume, weight and total circulating protein

and a three stage thoracoplasty was done under local anaesthesia. The first operation was performed in September, 1947, when the hyperalimentation program ended after nearly six months. The third stage was completed November 24, 1947. The patient tolerated the

surgical procedures very well. Following operation he slowly gained weight (11.5 lbs.) up to 108.5 lbs. on March 12, 1948 when the control period with the reinstituted regular diet terminated. Reference to table 6 and figure 18 indicates that the blood volume and total circulating protein followed in rather close parallel direction with the recorded weights.

In brief, it is seen that a loss of weight occurred with the hyperalimentation regimen and this trend was reversed after thoracoplasty when the regular diet was resumed.

COMMENT

The value of maintaining an optimum nutritional standard, aside from an adequate vitamin and mineral intake, in the treatment of a chronic disease, such as pulmonary tuberculosis, needs no defense. Unquestionably sufficient calories must be available to supply energy for basal needs and for tissue repair. While calories can be furnished by many foods, protein is an essential constituent of tissue which cannot be replaced by other foodstuffs. Furthermore, the storage of protein in the body is limited and must be constantly replenished in contrast, for example, to the body store of food in the form of adipose tissue.

The conventional quota of one Gm. of protein per Kg. body weight may suffice for the diet of the average individual but in patients with loss of weight, wasting of muscle, proteinuria, serous cavity exudates, et cetera, the allowance of protein must be increased. Lund and Levenson (3) and Elman (4) among many others have emphasized the manifestations of protein deprivation and its importance in relation to shock, susceptibility to infection, wound healing, nutritional edema, and in the hepatic detoxication of various toxins.

However, protein in the form in which it is ingested cannot be incorporated in the tissues. It must first be converted into amino acids which in turn become the basic constituents for the formation of the complex proteins of the cells and plasma proteins. This synthesis takes place primarily in the liver. It is possible that in some instances of tuberculosis a damaged liver may not carry out the normal function of manufacturing the proteins that it ordinarily furnishes to the body and which in turn protect it against further injury. It is interesting to note in 3 patients of the present series, that restoration to normal of an abnormal retention of the bromsulfalein dye followed the hyperalimentation regimen. It would be difficult to state, however, that these results by themselves may be used as an index of improvement in the protein metabolism of the liver.

The study of diet with the amino acids as tools has been projected in recent years for the correction of protein deficiency. Cannon (5) has postulated that for "tissue synthesis all the essential amino acids must be available in adequate amounts and proportions one to another and at approximately the same time." The employment of protein hydrolysates (by oral, tube feedings, or parenteral administration) as substitutes or supplements to the diet has become the vogue in the treatment of some medical and surgical conditions. Co Tui (6) commented on the oral use of a protein hydrolysate with Dextri-Maltose in 3 cases of advanced tuberculosis treated on the Chest Service of Bellevue Hospital. All showed an increase in weight and regression of disease and the latter became ap-

parent after five to seven months of therapy. It was felt that these patients revealed more improvement with the combination of bed rest and hyperalimentation than could have been expected ordinarily. In none, however, was there an increase in the plasma protein to match the increase in weight.

Many patients with advanced disease will show significant constitutional and roentgenologic improvement for variable periods of time with bed rest and the usual dietary program. In this report, the clinical proctocols of the patients, together with their photographs and chest roentgenograms at the time the hyperalimentation commenced, give adequate visualization of their generally poor condition and the far advanced tuberculosis. It is believed that the control periods of the experiment in these 6 cases in comparison with results obtained during the dietary therapy permitted a fair evaluation of hyperalimentation. Three patients (Cases I, III, IV) showed significant constitutional response with gain in weight for which the supplemental protein hydrolysate regimen deserves most or all of the credit. The pulmonary lesion was improved in 2 and stationary in one but these effects could not be properly attributed to hyperalimentation per se. One patient (Case I) died suddenly and 2 are living after the regular diet was resumed. One of these patients (Case III) lost weight and the other (Case IV) continued to increase his weight. The second group of 3 patients (Cases II, V, VI) lost weight during hyperalimentation and also revealed roentgenologic progression of tuberculosis, although in one the disease subsequently became quiescent probably due to streptomycin therapy, and thoracoplasy was performed. This patient (Case VI) thereafter gained weight on the regular diet alone. Another living patient (Case V) continued to lose weight after the ordinary hospital diet was reinstituted, while Case II died from further dissemination of pulmonary tuberculosis.

The laboratory test which has been used most widely in the detection of protein deficiency is the concentration of the plasma or serum protein. According to Peters and Eisenman (7) the normal range of total protein is from 6.0 to 8.0 Gm. per 100 cc.; the albumin is between 4.0 and 5.5 and the globulin from 1.4 to 3.0 Gm per 100 cc. This test, however, does not necessarily afford an index to the protein store in the body, as in patients with a slow development of protein malnutrition there may be no manifest hypoproteinemia until a considerable protein deficiency occurs in the tissues. Also in protracted infections there is frequently a shrinkage of the blood volume with loss of water which results in a relative increase in the serum protein (8). The actual concentrations of the albumin and globulin fractions are of more significance than their ratio. A lowered albumin, especially below 2.5 Gm. per 100 cc., is usually associated with frank edema although such factors as bed rest and dehydration may retard or lessen the edema. None of the patients actually showed edema or hypoproteinemia below the critical level; the globulin on occasions was high normal or above. Determinations of the plasma and blood volumes and the total circulating blood protein may give a more accurate picture of the state of protein nutrition, particularly in the earlier stages of deficiency and before the onset of edema. Chow (9) has stated that regeneration of total protein may occur after the administra-

tion of protein hydrolysate, even with a decrease of plasma protein concentration, because of a larger increase in plasma volume. In the present dietary study, the changes in the serial determinations of the circulating protein generally, though not invariably, were more significant and revealed closer correlation with the clinical course than did the variations in the serum protein concentration. In 3 cases also there was good parallelism between the blood volume figures and the weight records (Cases I, III, VI).

There is no doubt that large amounts of protein nourishment can be administered with hydrolysates and that in some patients with far advanced tuberculosis and significant weight loss a protein deficiency pattern can be favorably modified. Such patients are also frequently anemic and blood transfusions are an asset. Mixtures of amino acids will not improve the anemia at first, although they most likely aid later in the elaboration of hemoglobin. Transfusion therapy was omitted from this study so that the hyperalimentation program per se could be properly appraised. It was not the intention to undertake a therapeutic comparison of the hydrolysate preparations. Their use entails some objectionable features. Four patients (Cases II, III, IV, V) had gastro-intestinal disturbances of nausea, vomiting and/or diarrhea which in only 2 cases, however, were of moderate severity. The earlier experience also indicated that in 2 patients with frank symptoms of associated intestinal tuberculosis, the oral use of hydrolysates aggravated these symptoms. It is probable that the above untoward manifestations may be lessened by gradually increasing the dose of the hydrolysate up to the required level rather than starting immediately with the total prescribed quantity. The unpleasant taste of these products is a personal equation and can be masked somewhat by the addition of peppermint water. Most of the patients commented at times about the monotony of hydrolysate therapy and experienced some loss of appetite for natural foods but these complaints were not presented as serious disadvantages.

A basic question is whether the normal intake of a natural high protein diet may not accomplish what the hyperalimentation method sets out to do. The writers' past experience, though limited, has indicated that in tuberculous patients of poor general condition and extensive disease, the prolonged use of a diet containing 150 Gm. or more of whole protein is frequently tolerated with difficulty as the bulk is too great and the sense of fullness and satiation too discomforting. As mentioned previously, much more protein and in smaller bulk can be supplied in the hydrolyzed form than as whole protein. Furthermore, the hydrolysates provide a more uniform composition and their intake can be calculated more readily. Another advantage is that the hydrolysates spare the need for protein digestion. No direct evidence is available, however, to prove that in tuberculosis there is a failure to break down proteins into amino acids; significant enzymic dysfunction of the gastric and pancreatic juices has not been demonstrated. It therefore appears probable that protein concentrates, unhydrolyzed preparations, such as casein, wheat germ, soy bean flour or commercially available processed proteins, can be of value. These products have the added advantages of increased palatability, economy and lessened gastro-intestinal irritation.

SUMMARY

Protein hydrolysates containing varying amounts of amino acids were used orally to supplement the regular hospital diet in 6 tuberculous patients who had shown an unfavorable clinical or roentgenological course. The dietary therapy was continued for six to eleven months. Four of the surviving patients were then returned to the routine diet and were observed during a six month control period.

During hyperalimentation, 3 patients gained weight and the beneficial effect was attributed primarily to the therapy. The pulmonary lesions of these patients were either improved or stationary, as determined by roentgenography. One patient died suddenly. Of the two living patients, one lost and the other continued to gain weight after the regular diet alone was resumed. The remaining 3 patients (one died) lost weight during hydrolysate therapy and showed evidence of progression of their tuberculosis. After resumption of the regular diet, one patient lost and the other gained weight.

Tuberculous patients who show evidence of clinical deterioration, or in whom it may be desirable to attempt improvement of the nutritional status preliminary to or after thoracic surgery, may be suitable subjects for hyperalimentation with particular emphasis on increased protein. The advantages and disadvantages of the oral use of protein hydrolysates are discussed.

SUMARIO

La Proteohidrolisatoterapia Oral Complementaria en la Tuberculosis

Empleáronse por vía bucal hidrolisatos de proteína que contenían distintas cantidades de amino-ácidos esenciales para complementar la habitual alimentación hospitalaria en 6 tuberculosos que habían mostrado una evolución clínica o roentgenológica desfavorable. La dietoterapia continuó de seis a once meses. Cuatro de los sobrevivientes fueron entonces puestos de nuevo a la dieta corriente y observados durante un período de comprobación de seis meses.

Durante la hiperalimentación, 3 enfermos aumentaron de peso, atribuyéndose el beneficio primordialmente al régimen empleado. Según determinó la radiografía, las lesiones pulmonares de estos enfermos o bien mejoraron o se estacionaron. Un enfermo murió repentinamente. De los dos vivos, uno perdió y el otro continuó ganando peso después de reanudarse la dieta regular sola. Los otros 3 enfermos (uno murió) perdieron peso durante la hidrolisatoterapia y revelaron signos de agravación de su tuberculosis.

Los tuberculosos que revelan signos de deterioro clínico, o en los que puede convenir tratar de mejorar el estado nutritivo antes o después de la cirugía torácica, pueden constituir sujetos apropiados para la hiperalimentación, haciendo en particular hincapié en un aumento de proteína.

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NINETY CONSECUTIVE CASES OF PULMONARY TUBERCULOSIS TREATED BY A COMBINATION OF STREPTOMYCIN AND PNEUMOTHORAX[1,2]

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INTRODUCTION

In this article 90 consecutive cases of pulmonary tuberculosis, treated at Fitzsimons General Hospital during the years 1946–1948 with a combination of pneumothorax and streptomycin, are presented. In a separate communication (1) the writers have attempted to evaluate another type of collapse procedure, pneumoperitoneum, in connection with streptomycin.

Because of the inadequate results frequently obtained following the use of streptomycin alone (2, 3, 4, 5), the writers became interested in combining temporary collapse measures with streptomycin therapy. The purposes of these additional measures are twofold: (*1*) to maintain closure of cavities and promote their healing; (*2*) to obtain closure of cavities that did not close or could not be expected to close on streptomycin alone.

It is considered to be extremely important that cavity closure be obtained as soon after the initiation of streptomycin therapy as possible because of the high percentage of cases in which drug-resistant strains of tubercle bacilli emerge (6). At the Fitzsimons General Hospital this phenomenon occurred in 36 per cent of a series of patients who received 2 Gm. of streptomycin daily and in 29 per cent of a group treated with 1 Gm. daily. The combined series totalled 361 patients.

If each step in the therapeutic plan be carefully thought out, succeeding measures will not be jeopardized by overzealous but ill-advised or untimely use of streptomycin. It has become obvious from experience at Fitzsimons General Hospital that it is most advantageous to use a short course of streptomycin as early in the treatment as possible, to be followed by other measures if this is not definitive. Otherwise, in the face of drug resistance, if surgery is required it must be performed without chemotherapeutic protection against spread of the infection (7).

The same criteria for selection of collapse procedures, as recommended by Rafferty (8), were used in the cases of the present study as in nonstreptomycin treated cases.

It is believed that the trend of treatment of cavitary lesions in pulmonary tuberculosis is changing toward the use of streptomycin in conjunction with some type of collapse measure, as demonstrated by the following evaluation of 90 cases treated by the combined method.

[1] From the Medical Service, Fitzsimons General Hospital, Denver, Colorado.

[2] Published with permission of The Surgeon General, U. S. Army, who assumes no responsibility for the opinions expressed or conclusions drawn by the authors.

PLAN OF INVESTIGATION

Criteria for the selection of cases: The series consisted of 90 consecutive cases who underwent treatment for pulmonary tuberculosis at Fitzsimons General Hospital during the years 1946–1948. No special selection was made and all of the cases available at the time for study were included. The majority of the patients in the series were young white males on active duty in the armed forces or recently discharged. It is realized that for accurate evaluation of the treatment for pulmonary tuberculosis long term studies are necessary and that the cases in this series represent for the most part a short term observation, averaging approximately ten months. This is a consequence of the fact that patients are hospitalized at Fitzsimons General Hospital for only a relatively short period before being discharged to Veterans Hospitals. It is believed that reporting the results of this study is justified at this time because significant improvement occurred in only a few months.

All patients were maintained on absolute bed rest or bed rest with lavatory privileges. This was taken into consideration in interpreting the results, and in the cases evaluated as "good" or "excellent" the results definitely surpassed those which would normally be expected on bed rest alone.

Streptomycin regimens: Streptomycin was administered in the following schedule of doses: Forty-nine patients received 1 Gm. per day divided into 2 daily doses intramuscularly. The total duration of therapy in these cases ranged between one and four months but the majority were treated for two or three months. Thirty-two patients received 2 Gm. per day divided into 5 daily doses intramuscularly. In general, this dose was administered for a total period of four months. In 9 cases, however, the duration of therapy varied from two to eight months. In addition to the intramuscularly administered streptomycin, 4 patients received the drug as an aerosol. There were 3 patients who received experimental doses of streptomycin, each respectively receiving 2 Gm. every three days; 2 Gm. every five days; 0.5 Gm. every day; for a total of 120 days. The results obtained in these cases compared favorably with those receiving the standard dosages.

Toxic Reactions: In 2 patients receiving 1 Gm. per day, streptomycin was discontinued because of severe dermatitis, but the period of treatment was considered to be adequate. On the 1 Gm. per day dosage, 5 additional patients had mild symptoms, 2 moderate and 40 had no symptoms. Of the 38 patients on 2 Gm. per day dosage, 18 had no symptoms, 12 had mild symptoms and 8 moderate symptoms. The 3 patients on miscellaneous dosages developed no toxic symptoms.

The aim was to obtain adequate pneumothoraces in all cases as prescribed by Rafferty (8) and Alexander (11). In no case was positive pressure or marginal pneumothorax given. Twelve patients received a closed intrapleural pneumonolysis and one patient received an open pneumonolysis. Ten of these were successful, resulting in a good to excellent collapse and 3 were unsuccessful but the pneumothorax was maintained.

Three patients received a concurrent pneumoperitoneum and 8 patients received a phrenemphraxis on the pneumothorax side.

Six patients received pneumothorax on the less involved side as a preparatory measure for thoracoplasty for the more diseased lung.

Three patients received bilateral pneumothorax.

Two patients were pregnant and had uncomplicated deliveries while under study.

Bronchoscopy was performed in 45 of the cases or 50 per cent. Indications for bronchoscopy were as prescribed by McIndoe, Steele, *et al.* (12), and Sharp and Gorham (24).

The pneumothoraces were graded as "excellent," "good," "fair," and "poor." A pneumothorax was considered "excellent" if no adhesions were present and an adequate collapse was maintained. If a small adhesion was present that did not interfere with the collapse, the pneumothorax was considered as "good." In those cases where an apparently adequate pneumothorax was present in spite of adhesions the pneumothorax was considered "fair." The classification as a "poor" pneumothorax is self-explanatory.

Twenty-two pneumothoraces were classified as "excellent," 34 as "good," 18 as "fair," and 16 as "poor."

The roentgenographic results were classified into five categories. These were "excellent," "good," "fair," "slight," "no change or worse." A spectacular roentgenographic clearing plus cavity closure was graded as "excellent." A grade of "good" was given if marked roentgenographic regression occurred that was superior to that expected on bed rest but where cavity closure was not as prompt or as complete as in the "excellent" cases. Results which were better than normally anticipated on bed rest were classified as "fair." The remaining two categories, "slight" or "no change" are self-explanatory.

The grading of improvement in clinical symptoms closely follows that used for the classification of roentgenographic regression. Cough, sputum production, weight, toxicity, and erythrocyte sedimentation rate were the factors taken into consideration. Roentgenograms, sputum examinations, and erythrocyte sedimentation rates and weight determinations were obtained at monthly intervals. Cultures of the gastric washings and special roentgenograms were obtained whenever indicated.

There was only one minimal case of pulmonary tuberculosis in the series. Of the cases, 54 or 60 per cent were moderately advanced and 35 or 39 per cent were far advanced. The average period of observation was approximately ten months, the shortest being four

TABLE 1

Combined Treatment with Streptomycin and Pneumothorax

	TOTAL	SEX		COLOR			AGE				N. T. A. CLASSIFICATION		
		Male	Female	White	Colored	Others	Teen to 19	20 to 29	30 to 39	Over 40	Min.	Mod. Adv.	Far Adv.
Number.....	90	84	6	76	11	3	20	49	16	5	1	54	35
Percentage.....	100	93	7	85	12	3	22	54	18	6	1	60	39

TABLE 2

Combined Treatment with Streptomycin and Pneumothorax

	STREPTOMYCIN FOLLOWED BY PNEUMOTHORAX	STREPTOMYCIN AND PNEUMOTHORAX SIMULTANEOUSLY	PNEUMOTHORAX FOLLOWED BY STREPTOMYCIN
Total.....	50	15	25
Percentage.....	55	17	28

months and the longest four years. Every effort was made to be extremely conservative in the evaluation of these cases.

The efficacy of streptomycin in the treatment of moderately advanced and far advanced cases of pulmonary tuberculosis has been well demonstrated by many investigators (3, 10, 13, 14 and 25).

Rapid spontaneous regression was not likely to occur in these patients as they had received a course of bed rest therapy without regression prior to the institution of combined treatment.

Of the 90 patients treated, 76 were white, 11 were Negroes, and 3 were Chinese or Japanese. Eighty-four were males and 6 were females. The largest age group was between 20 and 29 which included 49 of the cases, and 20 were in their teens. Sixteen were between the ages of 30 and 39. Only 5 of the patients were over 40.

The series is divided into 3 groups. The first and largest group consisting of 50 cases, or 55 per cent, are those cases treated with streptomycin for at least one month prior to the induction of the pneumothorax. The second group consists of 15 cases, or 17 per cent, which had pneumothorax and streptomycin started simultaneously. In all cases in this group

both modes of treatment were started within a few days of each other. Twenty-five, or twenty-eight per cent, constituted the third series in which pneumothorax preceded streptomycin by more than one month. This threefold classification was made to evaluate the different therapeutic results that could be obtained from the various combinations (table 2).

RESULTS

Streptomycin Therapy Instituted Prior to Pneumothorax (50 cases)

The patients who were treated with streptomycin before the induction of pneumothorax continued to receive the drug for several days to several months following the start of collapse therapy. In 34 of this group, reversal of infectiousness had occurred at the time the information for this article was collected. Tubercle bacilli remained present in the sputum of 8 of these patients while they received only streptomycin, but the bacilli disappeared following induction of pneumothorax. Sixteen, or 32 per cent, of the patients continued to discharge tubercle bacilli in the sputum (table 3A).

Cavity closure was observed in only 5 cases on streptomycin alone. Forty-five had visible cavitation on the roentgenogram before pneumothorax was induced and in 19 of these patients the cavities closed following collapse therapy. Cavity closure occurred four times as frequently after pneumothorax was induced as while streptomycin therapy was used alone.

Cavities persisted in 26 patients after combined therapy, and the discharge of tubercle bacilli in the sputum continued in 16. In the group in which cavity closure was obtained, it was found that reversal of infectiousness frequently preceded the cavity closure by one to three months.

In Table 3B it may be seen that 11 of the 16 patients without "sputum conversion" have been followed for less than four months and that 20 of 26 patients who failed to experience cavity closure have also been followed for less than four months after institution of combined treatment. A further review at the end of six months of combined treatment would be especially valuable as Bendove *et al.* (15) show that 90 per cent of 546 cases becoming negative after pneumothorax had been induced did so by the end of the first semester.

Further comparison of the cases in which cavity closure occurred with those in which cavities persisted reveals that in the former group 18 had only moderately advanced lesions and 6 had far advanced lesions. In the group without cavity closure 13 cases were moderately advanced and 13 were far advanced. The ratio of moderately advanced to far advanced was 3 to 1 in the more successful group as compared to a 1 to 1 ratio in the less successful group. In retrospect the preferred type of treatment in several of these cases of far advanced disease without cavity closure may have been pneumoperitoneum rather than pneumothorax. Moreover, 6 of the cases without cavity closure were being prepared for thoracoplasty. The pneumothorax was maintained on the less diseased side and could not be expected to have any effect on the contralateral cavitary disease.

A total evaluation of this group reveals that 36, or 72 per cent, had either "good" or "excellent" roentgenographic improvement and that 32 or 64 per

cent had "good" or "excellent" clinical improvement. Only 3 patients showed no roentgenographic improvement or became worse and only one case failed to show any clinical improvement.

TABLE 3A

Streptomycin Instituted Prior to Pneumothorax
Reversal of Infectiousness and Cavity Closure

Before Combined Treatment

"Sputum conversion" on streptomycin alone	26 (52%)	Cavity closure on streptomycin alone	5 (10%)
No "sputum conversion" on streptomycin alone	24 (48%)	No cavity closure on streptomycin alone	45 (90%)
Total	50 (100%)	Total	50 (100%)

After Combined Treatment

"Sputum conversion" after pneumothorax added	34 (68%)	Cavity closure after pneumothorax added	24 (48%)
No "sputum conversion" after pneumothorax added	16 (32%)	No cavity closure after pneumothorax added	26 (52%)
Total	50 (100%)	Total	50 (100%)

TABLE 3B

Streptomycin Instituted Prior to Pneumothorax
Duration of Follow-up after Combined Treatment in Patients Without "Sputum Conversion" or Cavity Closure

	TOTAL	FOLLOW-UP—NUMBER OF MONTHS				
		1 to 2	2 to 3	3 to 4	4 to 6	6 to 9
No "Sputum Conversion"	16	0	6	5	2	3
No Cavity Closure	26	6	8	6	4	2

TABLE 3C

Streptomycin Instituted Prior to Pneumothorax
Cavity Closure

	TOTAL	MODERATELY ADVANCED		FAR ADVANCED		MORE THAN 50 PER CENT	LESS THAN 50 PER CENT
		Number	Per cent	Number	Per cent	Decrease in Cavity Size	Decrease in Cavity Size
Cavity Closure	24	18	75	6	25	Complete	Complete
No Cavity Closure after Combined Treatment	26	13	50	13	50	13 (50)	13 (50)

A comparison is shown in Table 4B between the adequacy of pneumothorax and the final results. It can be seen that there is a direct correlation between

the type of pneumothorax and the results as would normally be expected. Thirty-eight of the 50 cases showed "excellent" or "good" final results. Of these 29 had "good" or "excellent" pneumothoraces and 6 had a "fair" collapse.

Streptomycin and Pneumothorax Started Simultaneously

Pneumothorax and streptomycin were started at the same time in 15 cases. In 11 of these patients, tubercle bacilli disappeared from the sputum and in 10 cavity closure occurred. Of the 4 in whom "sputum conversion" did not

TABLE 4A

Group I: Streptomycin Instituted Prior to Pneumothorax
Total Evaluation at End of Observation Period

	TOTAL	"EXCELLENT"	"GOOD"	"FAIR"	"SLIGHT"	"NONE" OR "WORSE"
Roentgenographic Improvement	50	13 (26%)	23 (46%)	10 (20%)	1 (2%)	3 (6%)
Clinical Improvement	50	8 (16%)	24 (48%)	13 (26%)	4 (8%)	1 (2%)*

* One patient asymptomatic at start of therapy.

TABLE 4B

Group I: Streptomycin Instituted Prior to Pneumothorax
Final Evaluation of Results Correlated with Adequacy of Pneumothorax

FINAL RESULTS	NUMBER	PER CENT	TYPE OF PNEUMOTHORAX			
			"Excellent"	"Good"	"Fair"	"Poor"
"Excellent"	16	32	6	5	4	1
"Good"	22	44	2	16	2	2
"Fair"	7	14	2	2	2	1
"Slight"	2	4	0	0	2	0
"None" or "Worse"	3	6	0	0	1	2
Total	50	100	10	23	11	6

occur, 3 were followed for less than 4 months. The same short period of observation applied to 4 of the 5 patients in whom cavity closure did not occur. Twelve of the 15 cases, or 80 per cent, experienced either "excellent" or "good" roentgenographic results and 10, or 67 per cent, showed "excellent" or "good" clinical improvement (table 5B).

In the total evaluation of these cases, 9 were considered as having attained "excellent" results; 3, "good" results; 2, "fair" results, and one became worse.

Pneumothorax followed by Streptomycin

Twenty-five patients received pneumothorax prior to the start of streptomycin therapy. This series is comprised of two main groups: (*1*) those cases in whom pneumothorax alone had not produced adequate therapeutic results, and (*2*)

those cases in which streptomycin was added shortly after pneumothorax was induced. In all of these cases pneumothorax was continued after streptomycin was given.

Of the 25 cases, "sputum conversion" occurred in 5 on pneumothorax alone. In an additional 13 cases the sputum became negative for tubercle bacilli (culture) after streptomycin was added. In only 6 cases did the sputum remain positive for tubercle bacilli. A direct parallel can be drawn in regard to cavity closure. On pneumothorax alone, only 5 cases showed cavity closure on the roentgenogram, whereas an additional 12 had closure after streptomycin was

TABLE 5A

Streptomycin and Pneumothorax Started Simultaneously (15 cases)
"Sputum Conversion" and Cavity Closure

	NUMBER	PER CENT
"Sputum Conversion"		
Yes	11	73
No	4	27
Total	15	100
Cavity Closure		
Yes	10	67
No	5	33
Total	15	100

TABLE 5B

Streptomycin and Pneumothorax Started Simultaneously
Total Evaluation at End of Observation Period

	TOTAL	"EXCELLENT"	"GOOD"	"FAIR"	"SLIGHT"	"NONE" OR "WORSE"
Roentgenographic Improvement	15	6	6	2	1	0
Clinical Improvement	15	5	5	4	1	0

added. In 8 cases, visible cavitation persisted, but 4 of these have been followed for less than four months since combined therapy was instituted (table 6A).

Roentgenographic improvement was "excellent" in 5 cases, "good" in 11 cases, and 8 showed "fair" improvement. Only one case showed "slight" improvement and none became "worse".

A final, complete evaluation revealed that "excellent" clinical improvement occurred in 6 patients and 8 were classified as "good." Eight patients showed a "fair" improvement and 3 failed to improve (table 6B).

Nine patients in this group had what was considered an "excellent" pneumothorax and in 7 the collapse was considered as "good". Of these 16 patients,

TABLE 6A

Pneumothorax Induced Prior to Streptomycin Therapy
"Sputum Conversion" and Cavity Closure

Before Combined Treatment

"Sputum conversion" on pneumothorax alone	5 (20%)	Cavity closure on pneumothorax alone	5 (20%)
No "sputum conversion" on pneumothorax alone	19* (76%)	No cavity closure on pneumothorax alone	20 (80%)
Total	25 (100%)	Total	25 (100%)

After Combined Treatment

"Sputum conversion" after addition of streptomycin	18 (72%)	Cavity closure after addition of streptomycin	17 (68%)
No "sputum conversion" after addition of streptomycin	6* (24%)	No cavity closure after addition of streptomycin	8 (32%)
Total	25 (100%)	Total	25 (100%)

* In one patient with cavity, sputum and gastric washings were negative for tubercle bacilli throughout entire period of observation.

TABLE 6B

Pneumothorax Induced Prior to Streptomycin
Total Evaluation at End of Observation Period

	TOTAL	"EXCELLENT"	"GOOD"	"FAIR"	"SLIGHT"	"NONE" OR "WORSE"
Roentgenographic Improvement	25	5	11	9	0	0
Clinical Improvement	25	6	8	8	3	0

TABLE 6C

Pneumothorax Induced Prior to Streptomycin
Final Evaluation of Results Correlated with Adequacy of Pneumothorax

FINAL RESULTS	NUMBER	PER CENT	TYPE OF PNEUMOTHORAX			
			"Excellent"	"Good"	"Fair"	"Poor"
"Excellent"	11	44	4	5	2	—
"Good"	9	36	3	2	1	3
"Fair"	2	8	1	—	1	—
"Slight"	2	8	1	—	—	1
"Poor"	1	4	—	—	—	1*
Total	25	100	9	7	4	5

* Patient was AWOL frequently.

9 had "excellent" and 5 had "good" final results which also shows a clear correlation between results and the type of pneumothorax. In summary, 20 of the 25 patients, or 80 per cent, showed "good" or "excellent" final results when streptomycin was added to pneumothorax (table 6C).

TABLE 7

Bronchoscopy (45 of 90 cases)

	STREPTOMYCIN PRIOR TO PNEUMOTHORAX	STREPTOMYCIN AND PNEUMOTHORAX SIMULTANEOUSLY	PNEUMOTHORAX PRIOR TO STREPTOMYCIN	TOTAL
Total Bronchoscoped	27	2	16	45 (100%)
Endobronchial Disease				
No	14	1	8	23 (51%)
Yes	13	1	8	22 (49%)
Cured	10	—	8	18 (82%)
Improved	2	—	—	2 (9%)
Unchanged	1	1	—	2 (9%)

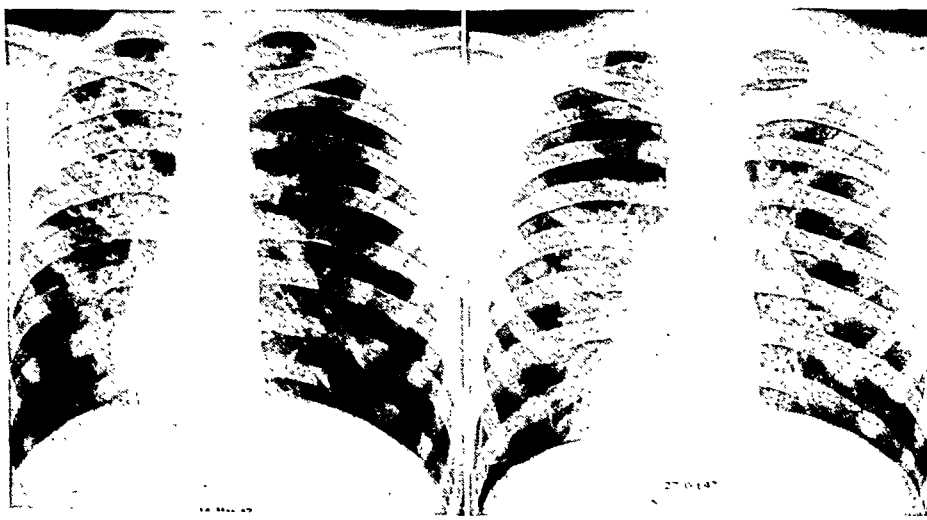

FIG. 1a. (Left) Case 1(a) before combined treatment; duration of disease for three months. Three large cavities are present in left upper lung.

FIG. 1b. (Right) Case 1(b) seven months after institution of simultaneous treatment with streptomycin (2 Gm. per day for 120 days) and left pneumothorax. The cavitation is no longer visible and there is marked clearing of the infiltration in the left lung. Last sputum positive for tubercle bacilli was in May, 1947.

BRONCHOSCOPY

Since 45 patients were bronchoscoped in this series, it is considered worth while to devote a paragraph to the findings. In 22 of the 45, or approximately 50 per cent of the cases, the typical findings of endobronchial tuberculosis were found. Three categories of endobronchial tuberculosis were designated: (*1*) submucosal, with a yellow productive nodularity on a reddened mucosa; (*2*)

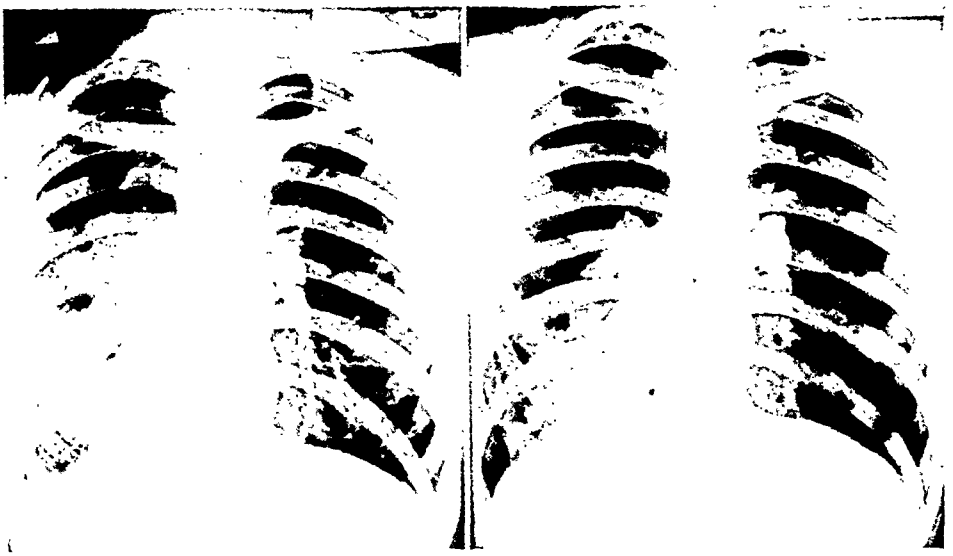

FIG. 2a. (Left) Case 2(a) before combined treatment; duration of disease for four months. Massive involvement of lower two-thirds of left lung with 3 cm. cavity in midlung field and a 3 cm. cavity in left hilar region.

FIG. 2b. (Right) Case 2(b) seven months after institution of combined therapy; left pneumothorax induced January 13, 1947. Streptomycin started February 17, 1947 (2 Gm. per day for 120 days). Last sputum positive for tubercle bacilli was in May, 1947.

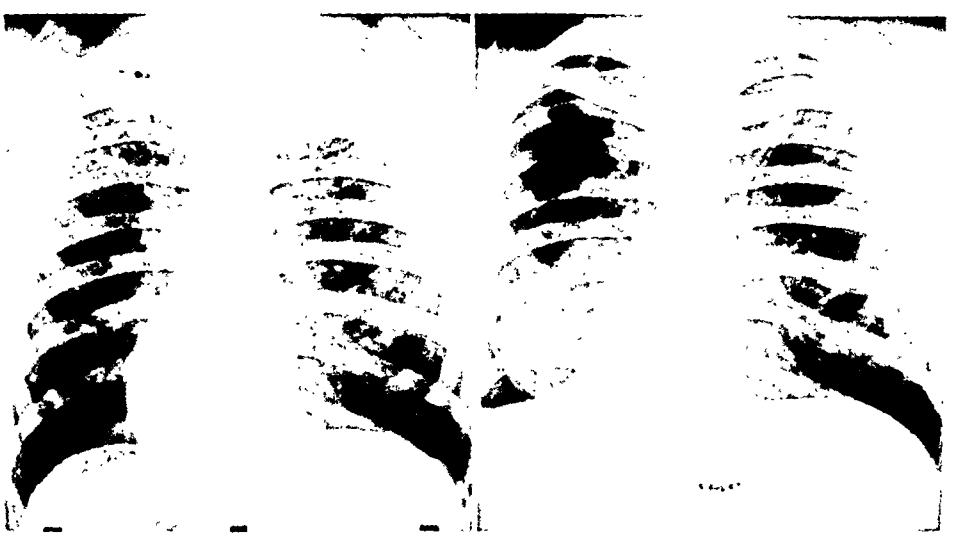

FIG. 3a. (Left) Case 3(a) before combined treatment; duration of disease for three months. Bilateral exudative-productive disease present involving all lobes of the left lung and right upper lung; 3 cm. cavity in left upper lung.

FIG. 3b. (Right) Case 3(b) seven months after institution of simultaneous treatment with streptomycin (2 Gm. per day for 150 days) and left pneumothorax. The cavity is no longer visible in the left upper lung and there is almost complete clearing of both lung fields. Last sputum positive for tubercle bacilli was in April, 1947.

ulcerative, showing a velvety, granular base with ulceration; and (3) fibrostenotic. The diagnosis was usually made on the basis of one examination. After treatment with streptomycin, 18 of the 22 patients, or 82 per cent, showed no evidence of endobronchial disease on bronchoscopy and 2 showed healing lesions. In two instances the lesions were unchanged as determined by bronchoscopy. The majority of the cases of endobronchial disease were present in the group which received streptomycin prior to pneumothorax. Thus the antibiotic treatment made it possible for an effective pneumothorax to be induced. In several other patients in whom tension cavities developed while receiving pneumothorax, streptomycin was instrumental in controlling the endobronchial tuberculosis and thereby promoting healing of the parenchymal lesion.

DISCUSSION

The writers are aware of the difficulty of evaluating the role played by either streptomycin or pneumothorax alone in the treatment of pulmonary tuberculosis and realize that it is even more difficult to attempt to evaluate the role played by each when they are used together. In addition cavity closure does not necessarily mean healing (7). It is believed, however, that by analyzing the results in this series certain deductions can be made.

It might be assumed when streptomycin was used first and followed by pneumothorax, that the therapeutic burden was carried by the antibiotic. The results seem to definitely indicate, however, that this is not the case. In the group in which streptomycin was used first, only 5 cavities closed on streptomycin alone. In an additional 19 cases, cavity closed roentgenologically after pneumothorax was added to the treatment, which represents a fourfold increase in closure. Other investigators who have used streptomycin alone in the treatment of cavitary disease report that it is not adequate treatment for obtaining permanent closure. D'Esopo (2) states "X-ray films of 11 of 14 patients revealed a reduction in size of cavities during the first 2 months of streptomycin therapy, but in only 1 instance did unequivocal cavity closure occur." In addition, the same writer and others have found that streptomycin-resistant tubercle bacilli appear in a high percentage of patients with persistent cavitary disease. It has also been the experience at Fitzsimons General Hospital, as well as that of other investigators, that a significant number of cavity closures occurring on streptomycin alone reopen after the streptomycin is discontinued (2). Therefore the writers are in agreement with D'Esopo (2) who believes "That there might be an optimum time during the course of streptomycin therapy when the institution of collapse procedures might have preserved what improvement had occurred."

Although pneumothorax itself is an accepted method of treatment for cavitary disease in pulmonary tuberculosis, as is borne out by numerous investigators (16, 17), this series demonstrates that in many cases pneumothorax produced optimum results only after streptomycin had been added. This was shown by the group of 25 pneumothorax cases in which streptomycin was given as an adjunct. Only 5 of these cases experienced cavity closure on pneumothorax alone, whereas

an additional 12 experienced cavity closure when streptomycin was added. A strikingly comparable result was seen in the reversal of infectiousness.

Nine of the 18 cases which eventually showed "sputum conversion," had been maintained on pneumothorax from four to twelve months prior to the addition of streptomycin therapy. Eight of these 9 "converted" their sputum within the first two months after streptomycin was added to the pneumothorax. This indicates the role of the combined treatment in hastening the healing of an otherwise sluggishly responding process.

Excellent results were also obtained in a smaller group in which the combination of streptomycin and pneumothorax was introduced simultaneously. Eleven of the 15 cases "converted" their sputum and 10 obtained cavity closure. This further bears out the efficacy of combined treatment.

Long term observations will be necessary to determine the optimum time either for introducing pneumothorax as an adjunct to streptomycin or streptomycin as an adjunct to pneumothorax in cavitary disease. Even at this early stage, however, certain factors can be recognized.

Pneumothorax has been shown to decrease the circulation of the collapsed lung by Dock (18). It is theoretically possible, therefore, that streptomycin should be given for several weeks prior to the induction of the pneumothorax in order to obtain maximum streptomycin concentration in the diseased tissue.

It appears probable that, if a pneumothorax is introduced while the patient is receiving streptomycin, a certain amount of protection against post-induction empyema and spread of disease is afforded. In the 65 cases presented in this series in which "protection" was given by streptomycin at the time of induction, no case of extension of disease or empyema occurred. In pre-streptomycin days, empyema usually occurred in 5 to 10 per cent of the cases in which pneumothorax was maintained and extension of the pulmonary disease occurred in an appreciable number. Gibbons (19) reports that 10.2 per cent of 663 cases developed empyema during pneumothorax treatment. The usual incidence of purulent effusion occurring during pneumothorax is discussed by Weisman (20). In 72 of 150 patients, pleural effusion was noted following the induction. Thirty-three effusions were aspirated of which 10 were originally purulent, and an additional 7 later became purulent. The peak incidence of effusions was reached early in the third month.

Streptomycin has been demonstrated to be most effective if introduced early in the course of the disease. The exudative stage of the lesion is most prominent at this time and this is known to respond best to streptomycin (21). This must be taken into consideration before inducing a pneumothorax alone, in that the exudative element will gradually comprise a smaller percentage of the pathological process as time elapses. If it is then found that a pneumothorax is incapable of healing the disease alone, introduction of streptomycin may not be as efficacious as it would have been much earlier in the disease process.

It seems to the authors that the above mentioned facts and data indicate that in the majority of cases the optimum treatment of pulmonary tuberculosis with cavitation will result from a course of streptomycin started several weeks before

the induction of the pneumothorax. The streptomycin should be continued for one to two weeks following the induction of the pneumothorax and in almost all cases the entire course of streptomycin therapy should comprise approximately six weeks of treatment at 1 Gm. per day intramuscularly (22).

In the entire series of 90 cases, 70 patients, or 78 per cent, had "excellent" or "good" final results. Of these, 50 had "excellent" or "good" pneumothoraces and an additional 11 were classified as "fair." Thus there is seen to be a very definite correlation between the adequacy of pneumothorax and the final therapeutic results. This in itself substantiates the value of pneumothorax in the combined method.

Not only can good final therapeutic results be obtained in a large percentage of cases, but a further type of application of the combined method is seen in a group of 6 cases of far advanced disease where prognosis had previously been hopeless. In each of these 6 cases a short course of streptomycin was given as well as pneumothorax. The combination was effective in preparing these cases for a thoracoplasty on the more involved side. The ultimate salvage of these cases is a tribute not only to streptomycin but to its judicious use in a well planned therapeutic approach to each individual case.

It has been the policy at Fitzsimons General Hospital to outline the tentative plan of treatment for each case of pulmonary tuberculosis. One of the first steps in the plan is a bronchoscopy. This was performed in 45 of the 90 cases and revealed a 50 per cent incidence of endobronchial tuberculosis. The presence of endobronchial tuberculosis acts as one of the determinants for the use of streptomycin as a first step in the plan of treatment. This then safeguards the subsequent induction of a pneumothorax (23).

In evaluating the end results of a planned method of treatment utilizing a combination of streptomycin and pneumothorax, the following significant data were obtained. Eighty-nine of the 90 cases had sputum positive for tubercle bacilli prior to therapy. Of these, 63 "converted." Cavity closure was attempted in 84 cases and 51 had successful roentgenographic closure. There were 6 other patients of the original 90 who did not receive pneumothorax on the side with the cavitary disease, but were being prepared for thoracoplasty by a contralateral pneumothorax. The average period of observation for the entire series was ten months. Although these 90 cases of combined treatment represent a short term study, it is believed that the results are significant and indicate a new trend in the therapy of pulmonary tuberculosis.

SUMMARY

The results of a study of ninety consecutive cases of pulmonary tuberculosis treated at Fitzsimons General Hospital from 1946 to 1948 with intramuscular streptomycin and pneumothorax are presented. One case of minimal pulmonary tuberculosis, 54 moderately advanced lesions and 35 far advanced cases were studied in the series. The results indicate that a combined method of treatment employing pneumothorax and streptomycin is a very effective means of treatment for pulmonary tuberculosis.

SUMARIO

Noventa Casos Consecutivos de Tuberculosis Pulmonar Tratados con una Combinación de Estreptomicina y Neumotórax

El estudio presentado versa sobre 90 casos consecutivos de tuberculosis pulmonar tratados en el Hospital General Fitzsimons de 1946 a 1948 con estreptomicina intramuscular y neumotórax. La serie comprendió un caso de tuberculosis pulmonar mínima, 54 lesiones moderadamente avanzadas y 35 casos muy avanzados. El resultado indica que un método terapéutico que combina el neumotórax y la estreptomicina ofrece un medio muy eficaz de tratamiento para la tuberculosis pulmonar.

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THE EFFECT OF STREPTOMYCIN, PARA-AMINOSALICYLIC ACID (PAS) AND THEIR COMBINATION ON THE TUBERCLE BACILLUS IN VITRO AND IN VIVO[1,2,3]

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(Received for publication June 16, 1948)

INTRODUCTION

Para-aminosalicylic acid (PAS) as a bacteriostatic agent against the tubercle bacillus was suggested by Lehmann (1, 2), who based his investigations on the work of Bernheim (3). The latter had observed that the sodium salts of benzoic and salicylic acids specifically stimulated the oxygen uptake of the organism. The tuberculostatic effect of PAS was quickly confirmed by Sievers (4) and by Youmans (5).

In a previous paper (6) a detailed comparison was made of the response of the tubercle bacillus *in vitro* to various concentrations of PAS, streptomycin, and the combination of these two drugs. It was found that PAS inhibited growth of both the streptomycin-sensitive strain, H-37RV, and the naturally occurring streptomycin-resistant variant, H-37RVNR1, almost completely in concentrations of 1.2 γ/cc. and partially in concentrations of 0.6 γ/cc. The inhibition of growth of H-37RV was markedly enhanced if a subinhibitory concentration of one drug was combined with a moderately inhibitory concentration of the other. This enhancement of inhibition by the combination of PAS and streptomycin was not found when it was used against the H-37RVNR1 strain. Another resistant strain, isolated from a patient following clinical treatment with streptomycin, a strain which was moderately inhibited by the highest concentration of streptomycin used (3700 γ/cc.) did, however, show the enhancement of inhibition when exposed to the combination of the two drugs.

METHODS

The *in vivo* experiments were carried out in guinea pigs. The virulent human strain of tubercle bacilli H-37RV was used to infect the animals. Each pig was inoculated subcutaneously with 0.1 mg. (wet weight) of a 2 week old culture of organisms grown in Dubos medium. Twenty-one days after infection the animals were divided into four groups and care was taken to ensure an equal distribution of animals of similar size and weight. The first group was treated with streptomycin alone, the second with PAS alone, the third with a combination of the two drugs, and the fourth was used as untreated controls. A portion of each of the first three groups was treated for 24 days and another for 45 days. A portion

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[2] Presented before the Medical Section, as part of the symposium on *Chemotherapeutics and Antibiotics*, at the 44th Meeting of the National Tuberculosis Association, New York, New York, June 16, 1948.

[3] This investigation was supported by a grant from the Division of Research Grants and Fellowships of the National Institute of Health, U. S. Public Health Service.

of the control group was sacrificed at the time treatment was started to determine the extent of disease at this time, while the other control animals were killed at the end of the two treatment periods. Of the treated animals the majority (46) were sacrificed after 24 days of treatment or died within near range of that period, while 18 animals were treated for 45 days or died shortly before the termination of the experiment. The exact arrangement of the experiment with divisions made according to the length of treatment is shown in table 1.

The stroptomycin was dissolved in sterile distilled water and was given in one daily dose of 8 mg. contained in 1 cc. of solution. The PAS was carefully weighed, dissolved in distilled water, brought to a pH of 7.0 with NaOH and sterilized by passage through a Seitz filter. It was prepared in a 2.5 per cent solution and a total daily dose of 250 mg. was given by subcutaneous administration of 5 cc. of the solution twice daily. The large dose of PAS

TABLE 1

Arrangement of Experiment

70 guinea pigs inoculated subcutaneously with 0.1 mg. of tubercle bacilli (H-37RV)

Treatment begun 21 days after inoculation

EXPERIMENT	NUMBER OF GUINEA PIGS	DRUG	ROUTE	TOTAL DAILY DOSE	DISTRIBUTION	NUMBER OF DAYS OF TREATMENT
Ia	10	streptomycin	subcutaneous	8 mg.	1 dose	24
Ib	16	PAS	subcutaneous	250 mg.	2 equal doses	24
Ic	10	streptomycin and PAS	subcutaneous	8 mg. 250 mg.	1 dose 2 doses	24
IIa	7	streptomycin	subcutaneous	8 mg.	1 dose	45
IIb	Animals intended for PAS treatment in this group died prematurely and were included in Ib.					
IIc	7	streptomycin and PAS	subcutaneous	8 mg. 250 mg.	1 dose 2 equal doses	45
			Controls			
IIIa	10	Killed at beginning of treatment				
IIIb	6	Killed with Group I				
IIIc	4	Killed with Group II				

used was decided upon after consideration of the 14 to 15 Gm. daily dose used in clinical trials (1, 2, 7, 8).

Careful postmortem examinations were performed on all animals. Sections of spleen, liver, and lung were taken and both hematoxylin and eosin and acid-fast stains were made. An evaluation of the amount of tuberculosis present in each animal was made from combined study of the macroscopic and histological findings.

RESULTS

The results of treatment as compared with the findings in the control groups are presented in figures 1, 2, and 3. It may be seen from figure 1 that systemic tuberculosis was present in all control animals at the time treatment was started. It is believed that a more reliable evaluation of the effect of chemotherapeutic agents in tuberculosis is obtained if treatment of the animals is withheld until the disease has been fully established.

In figure 2, which shows the results of the short-term therapy, it is obvious that

= Grade I = Grade III

= Grade II Grade IV

FIGS. 1, 2, and 3.—An arbitrary scale of four grades of tuberculous involvement evaluated from the macroscopic extent of disease with microscopic confirmation.

The numbers under some of the animals indicate that these died spontaneously; the numbers under control animals indicate number of days after infection when death occurred; the top numbers under treated animals indicate the number of days of treatment; the bottom numbers, the number of days after infection on which death occurred.

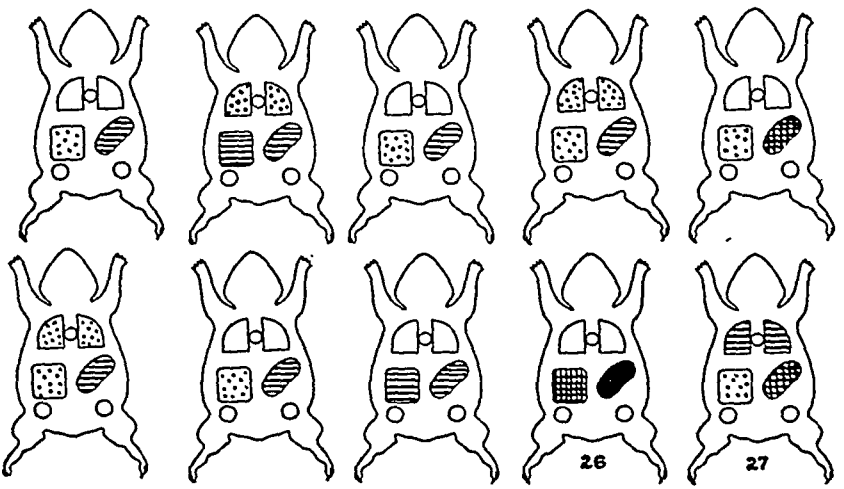

FIG. 1. Pre-treatment controls

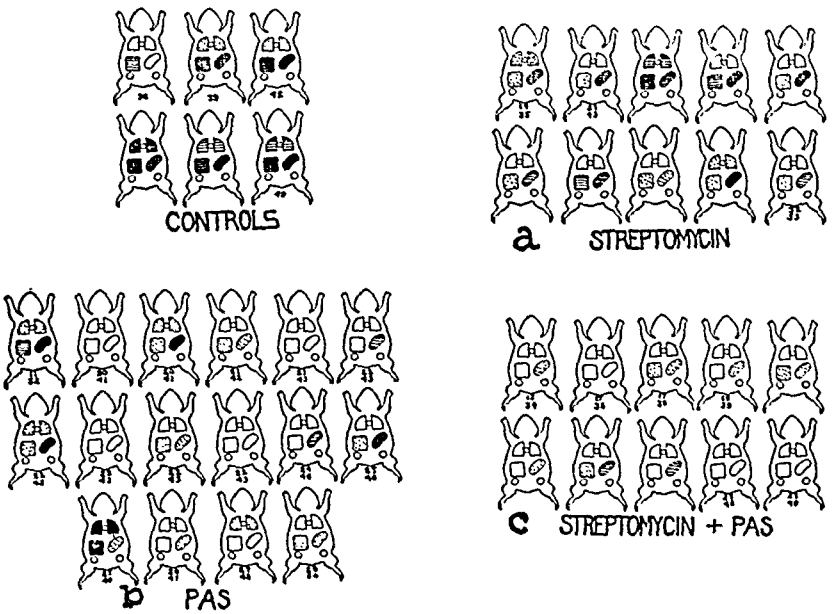

FIG. 2. Experiment I 24 day treatment

the tuberculosis in the untreated animals had continued to progress while those receiving streptomycin alone had on an average no more extensive disease than at the time treatment was started. The results in the group receiving PAS alone appear to be better than in the streptomycin group, but it must be remembered that the dose of PAS was very large (30 times that of streptomycin), so this does not indicate any superiority of PAS over streptomycin. The most striking results in the short-term treatment groups were found in the one receiving a combination of the two drugs. Only 2 animals in this group had as much as 2

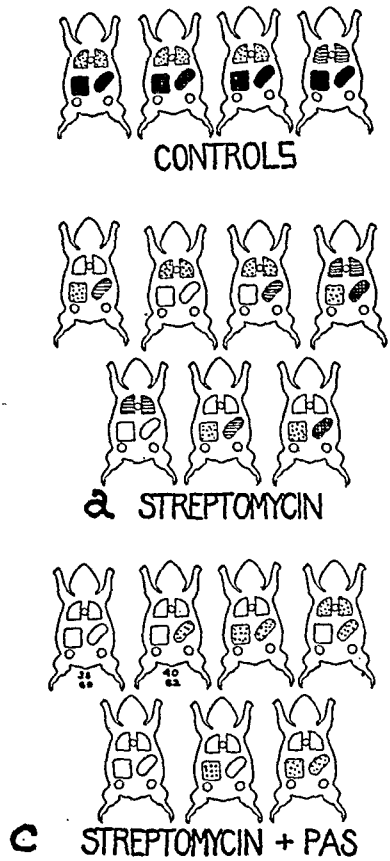

FIG. 3. Experiment II 45 day treatment

plus involvement of any organ, while the others had only minimal disease or none at all.

Figure 3 shows the results of the longer term of therapy. Again, the control animals which lived until the termination of the experiment showed further progression of the disease. Those receiving streptomycin for this length of time were beginning to show actual regression in the extent of disease, while those receiving both streptomycin and PAS showed a strikingly small amount of involvement which was no more than minimal in any of the animals.

The results of the complete experiment are expressed numerically in table 2,

which gives the average tuberculosis index for each of the groups. As in the culture experiments, the findings in animals indicate the striking inhibitory effect of the combination of the two drugs compared with that of each of the agents alone.

The microscopic as well as the macroscopic involvement of the lungs was comparatively slight even in the controls. The difference in the histologic changes between the treated groups of animals and the control groups was especially marked in the spleen and the liver.

In all the treated groups the changes were essentially the same except for the fact that they were most pronounced and frequent in the group receiving the combination of streptomycin and PAS. They consisted of two types of response. The first type was no different from the typical regressive changes seen in any case of healing tuberculosis, i.e., progressive fibrosis ending in complete disappearance of tuberculoid structure (figure 4a) and occasional calcification, especially in the spleen (figure 4b). The second type of response occurred chiefly

TABLE 2

Tuberculosis Index

Combined Macroscopic and Microscopic Findings

TREATMENT	KILLED AT BEGINNING OF TREATMENT	TREATED 24 DAYS OR DIED	TREATED 45 DAYS
Controls	4.4 (10)	7.5 (6)	8.5 (4)
Streptomycin		4.3 (10)	3.1 (7)
PAS		2.62 (16)	—
Streptomycin and PAS		1.2 (10)	1.14 (7)

The tuberculosis index is calculated from combined macroscopic and microscopic findings in the lung, liver, and spleen, grading each organ from 1 to 4. The maximum index possible is 12. The figures given are the average indices for each group. The figures in parentheses are the number of animals in the groups.

in the lungs and was similar to the changes reported by Krause (10) in anthracosis of guinea pigs. In some sections very large numbers of rounded lymphocytic infiltrates without a trace of specific reactions could be seen (figure 4c); it was impossible to decide whether all of them were residuals of pre-existing tubercles. In other sections the centers of similar structures appeared slightly lighter than the periphery so as almost to suggest germinative centers. When examined under high power, however, a number were recognized as tubercles in various phases of regression. At first the epithelioid cells showed a peculiar shrinkage, causing an early collapse of the typical architecture of the tubercle (figure 4d). At the same time a dense lymphocytic infiltrate formed at the periphery of the lesion. The center then gradually shrank to complete disappearance, leaving only a round accumulation of lymphocytes (figure 4e), with or without a few scattered giant cells. Eventually the lymphocytes too disappeared, and the only residual was a small streaky scar which was infiltrated with a few round cells. Occasionally in the liver a few giant cells singly or in small groups remained scattered without any surrounding granulation tissue (figure 4f).

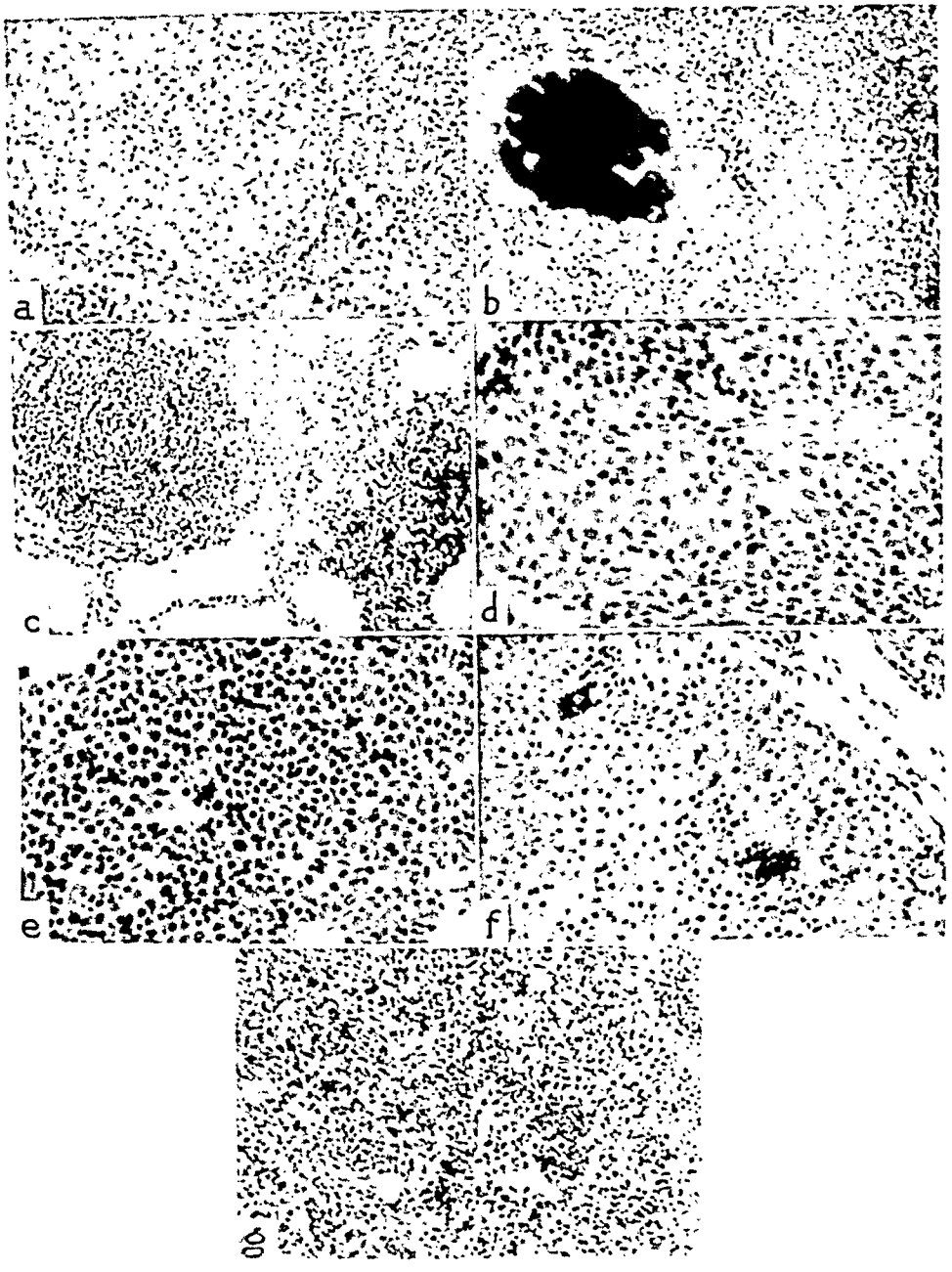

FIG. 4. a. Spleen, a completely fibrosed malpighian follicle. b. Calcification in the spleen. c. Lymphocytic accumulations in the lungs. d. Tubercle with small, shrunken epithelioid cells and peripheral lymphocytic infiltration. e. Further atrophy of epithelioid cells with complete disappearance of tuberculoid structure. f. Isolated giant cells in the liver. g. Myeloid metaplasia in spleen. Sinuses full of myeloid cells.

Another interesting finding was intense myeloid metaplasia in the spleen (figure 4g) and sometimes in the liver. Occasionally the blood in all vessels contained such large numbers of myeloid cells as are usually seen only in cases of true leukemia. Although this observation was made in 2 of the streptomycin-

treated pigs and even in one of the untreated controls, it was much more pronounced in the animals which had received PAS either alone or in combination with streptomycin. It was seen in a total of 12 such animals, 7 of which had received PAS alone and 5 the combined treatment.

The toxic effects of this large dose of PAS were considerable. Subcutaneous administration caused much local induration of the tissues and at autopsy hemorrhagic and necrotic areas were found at the sites of injection. No specific systemic lesions could be made out, but the majority of animals receiving PAS alone died before termination of the experiment. Premature death in almost all cases seemed to be due to pneumonia. It should be pointed out here that while Lehmann (2) found PAS to be highly toxic to the guinea pig, other workers (9) failed to find an excessive toxic effect from oral administration of the drug to this animal.

Smears and cultures were made from the spleens of some of the animals at the time of autopsy. Growth of tubercle bacilli in Dubos medium was obtained from 5 of these. Two had received streptomycin alone for 24 days, one had received streptomycin alone for 45 days, and 2 had received both streptomycin and PAS for 45 days. Preliminary studies on the streptomycin-sensitivity of these organisms showed that those isolated from the animals receiving the short-term streptomycin therapy were still sensitive to 0.74 γ streptomycin per cc. Those isolated from the animal receiving the long-term streptomycin therapy were resistant to 5.92 γ/cc., which was the highest concentration used in the preliminary run; while those from the 2 animals receiving the long-term combined therapy were still sensitive to 0.74 γ. Admittedly, this is too small a number of cases from which to draw any definite conclusions, but it does suggest that use of the combination of PAS and streptomycin may prevent the rapid appearance of resistant strains of tubercle bacilli.

DISCUSSION

On the basis of these *in vitro* and *in vivo* experiments with PAS and streptomycin in combination, two important clinical possibilities became apparent. First, it would seem feasible that, using a combination of the two drugs, smaller doses of streptomycin could be used effectively in the treatment of tuberculosis, thus reducing the hazard of toxic effects from this drug. Second, a delay in the emergence of streptomycin-resistance might be anticipated on theoretical grounds since PAS should be capable of suppressing growth of streptomycin-resistant strains. Insufficient evidence has been accumulated to support this view but it remains a promising possibility.

SUMMARY

1. Streptomycin alone, even in small doses given once a day and for a short period of time, will prevent the progression of tuberculosis in the guinea pig.

2. Para-aminosalicylic acid alone in large doses has a favorable effect on experimental tuberculosis.

3. The combination of streptomycin and PAS has a much more favorable effect on experimental tuberculosis in the guinea pig than either drug used alone.

SUMARIO

El Efecto de la Estreptomicina y el Acido Para-Aminosalicílico (PAS) y de la Combinación de Ambos sobre el Bacilo Tuberculoso in Vitro e in Vivo

1. La estreptomicina sola, aun a dosis pequeñas administradas una vez al dia y durante un breve período de tiempo, impide la agravación de la tuberculosis en el cobayo.

2. El ácido para-aminosalicílico solo a dosis masivas ejerce efecto favorable sobre la tuberculosis experimental.

3. La combinación de la estreptomicina y del PAS ejerce un efecto mucho más favorable sobre la tuberculosis experimental del cobayo que una u otra droga por sí sola.

Acknowledgment

The authors gratefully acknowledge the generosity of Parke, Davis & Company who supplied the para-aminosalicylic acid.

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THE EFFECT OF TUBERCLE BACILLI ON THE MIGRATION OF PHAGOCYTES IN VITRO[1]

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INTRODUCTION

In previous experiments (1), the fate of living virulent and avirulent tubercle bacilli injected into the peritoneal cavity of white mice has been studied. Observations made during the first hours of infection led to the conclusion that the virulent bacilli phagocytized by the polymorphonuclear leukocytes of the exudate cause injury to the leukocytes. This results eventually in lysis of the white cells with release of the phagocytized bacilli into the peritoneal fluid. The avirulent variants of tubercle bacilli, on the contrary, did not seem to cause any injury to the leukocytes which engulfed them. After being phagocytized, they are eliminated from the exudate.

These conclusions were drawn from observations made by examining peritoneal exudates. It is the purpose of the experiments reported in this paper to study the effect of phagocytized virulent and avirulent tubercle bacilli on leukocytes in a more direct way *in vitro*. The ability of the leukocytes to migrate was chosen as a test to demonstrate a possible injurious effect of engulfed tubercle bacilli on the leukocytes. Evidence is presented that the migration of polymorphonuclear leukocytes is completely inhibited by phagocytized virulent bacilli and unaffected by avirulent variants of the same strain when tested in comparable amounts.

METHODS

All the experiments referred to in this paper were performed with two variants of one strain of human tubercle bacilli, the virulent variant H37Rv, and the avirulent variant H37Ra[3] (2). Human leukocytes were used and prepared as follows: Blood from patients with high leukocytic counts (15,000 per cu. mm. or more) was defibrinated by gentle shaking in an Erlenmeyer flask. Two and six-tenth milliliters of the defibrinated blood were placed into small test tubes and 0.4 ml. of washed bacillary suspensions were added (0.4 ml. of Tyrode solution for the control tubes). The stoppered tubes were slowly rotated for twenty minutes at 37° C. and smears made to ascertain that phagocytosis had occurred. After short centrifugation at 1,500 r.p.m. most of the supernatant fluid was discarded, 0.2 ml. of fresh chicken plasma was added and thoroughly mixed with the blood. The tubes were then centrifuged for four minutes at 4° C. and allowed to stand for twenty minutes at room temperature. At this time the buffy coat had formed a solid membrane with the clotted chicken plasma and could be removed from the tube. After repeated washings in Ringers solution it was cut into small squares of 1 to 2 mm. side length. These squares were again washed in Ringers solution.

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[2] Present address: The Public Health Research Institute of the City of New York, Inc.

[3] These strains, received through the courtesy of Dr. R. J. Dubos, were originally obtained from the National Tuberculosis Association Standard Culture Depot at Trudeau, N. Y., and subcultured on Tween-albumin media.

Plasma, preferably from the same patient, was prevented from clotting by keeping it at low temperature and was diluted 2:1 with cold Tyrode solution and distributed in small Carrel flasks in amounts of 2.0 ml. One square piece of buffy coat was imbedded in the center of each flask and the plasma permitted to clot at room temperature. The flasks were incubated at 37° C. and the results observed twelve to fourteen hours later.

A few experiments were carried out with leukocytes from one day old peritoneal exudates of guinea pigs or rabbits and their corresponding plasma. The results were essentially identical with those obtained with human blood.

Leukocytes prepared in the way described above and containing no bacteria keep their characteristic tendency to migrate. At the very beginning of the incubation period, the first cells can already be observed migrating radially from the square piece of buffy coat. At the end of the incubation period a halo of 2 to 4 mm. in radius surrounds the original piece. The whole culture now forms a remarkably regular round disk of 5 to 8 mm. diameter. The halo consists chiefly of polymorphonuclear leukocytes. The size of the disk varies slightly from one blood donor (or animal) to the other, but remains fairly constant within one experiment (figure 1). Since there is a close macroscopic resemblance of this preparation to an ordinary tissue culture, it may be pointed out that the halo is not composed of newly grown cells but only of leukocytes emigrating from the original piece of buffy coat (figure 2).

RESULTS

The effect of virulent tubercle bacilli on the migration of leukocytes: When leukocytes were allowed to phagocytize virulent tubercle bacilli before being implanted, their migration was inhibited. The degree of the inhibitory effect depended upon the ratio bacteria/phagocytes (table 1). In preparations where, owing to the small number of bacilli, only part of the leukocytes had a chance to phagocytize bacilli, a slight migration still occurred. It could be seen, however, that all the leukocytes which contained bacilli did not leave the center of the preparation, and that all the leukocytes found in the surrounding halo were free from bacilli. With a higher number of bacteria, inhibition was complete (figure 3).

The effect of avirulent tubercle bacilli on the migration of leukocytes: In contrast to the effect of virulent bacilli, the avirulent variant did not inhibit migration. Cells with engulfed avirulent bacilli migrated as well as the controls (figure 4). Slight inhibition occurred only if a very high number of avirulent bacilli were added to the cell suspension so that almost all of the phagocytes contained many microorganisms (table 1). The rate of phagocytosis was the same with avirulent and virulent bacilli.

The effect of dead tubercle bacilli on phagocytes: Phagocytized heat-killed tubercle bacilli of both the virulent and the avirulent variants showed essentially the same effect on the migration of leukocytes as the living bacteria.

The effect of other phagocytized material on the migration of leukocytes: Control experiments were made to study the effect of charcoal, India ink and two strains of staphylococci on the migration of leukocytes. Phagocytized charcoal and India

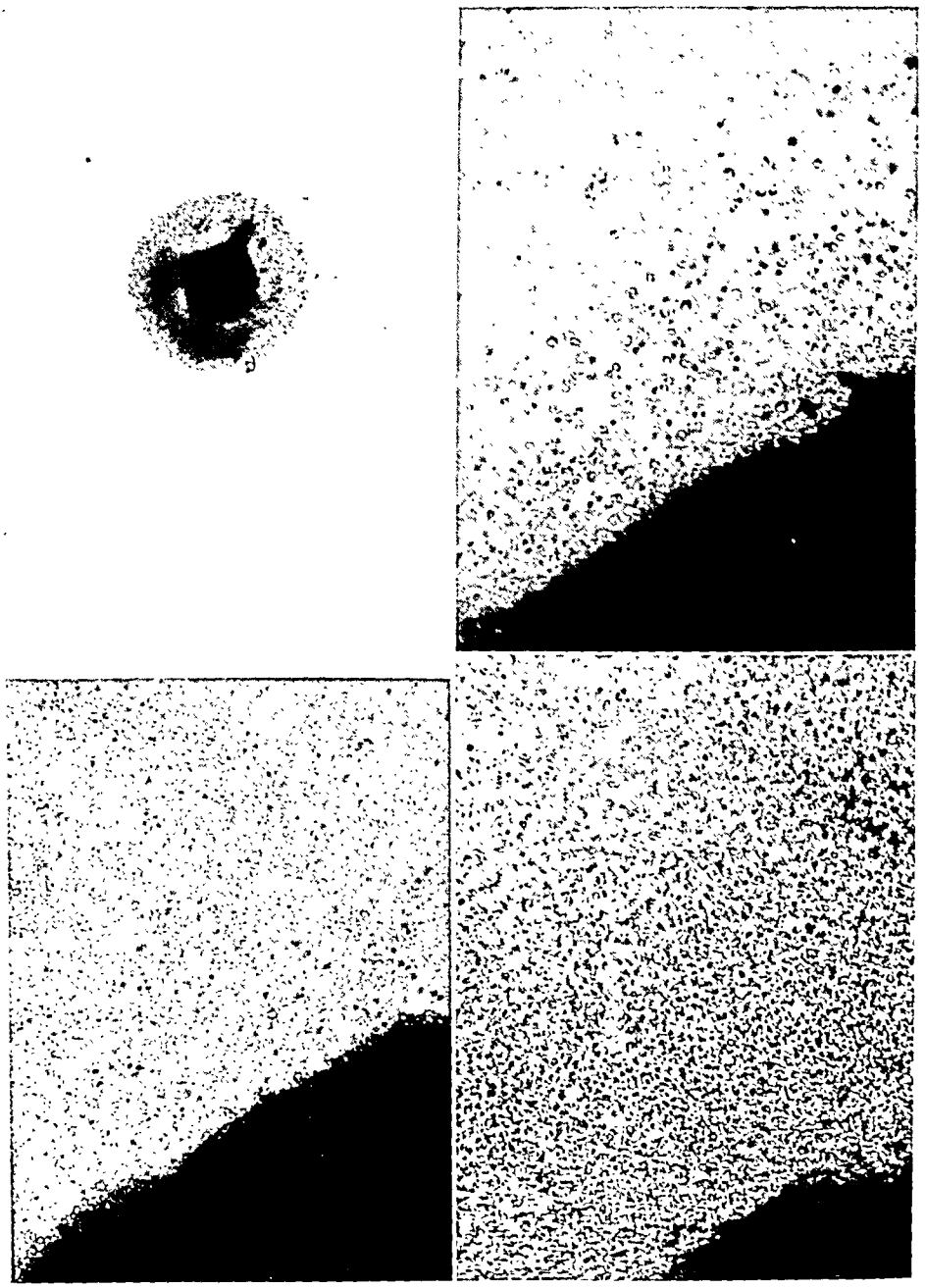

FIG. 1: (Upper left) The migration of human polymorphonuclear leukocytes after 14 hours incubation. No bacilli added. The figure shows the square piece of buffy coat imbedded in plasma, and the halo of leukocytes migrating from the original explantate. Magnification 10 ×.

FIG. 2: (Upper right) Section of a similar preparation as in figure 1 showing the edge of a young explantate with migrating leukocytes at a higher magnification (300 ×).

FIG. 3: (Lower left) The migration of human polymorphonuclear leukocytes after 14 hours incubation. Before being imbedded the leukocytes were allowed to phagocytize virulent tubercle bacilli; no migration occurred. (Magnification 300 ×).

FIG. 4: (Lower right) The migration of human polymorphonuclear leukocytes after 14 hours incubation. Before being imbedded the leukocytes were allowed to phagocytize avirulent tubercle bacilli; the migration of the cells is not inhibited by the engulfed bacilli. (Magnification 300 ×).

ink particles had no effect on the rate of migration, even when the phagocytes were heavily loaded. On the other hand, one freshly isolated strain of *St. aureus* was highly inhibitory to leukocytic migration, whereas an old laboratory strain did not affect the locomotion of the cells.

DISCUSSION

The results of these *in vitro* experiments show that virulent tubercle bacilli are injurious to polymorphonuclear leukocytes as evidenced by inhibition of the migration of the phagocytes. Since avirulent tubercle bacilli do not have a similar effect when tested in comparable number, it is likely that the inhibition of migration is in some way connected with the virulence of the bacilli. On the other hand, when a large number of avirulent tubercle bacilli is phagocytized, a slight

TABLE 1

Effect of Virulent and Avirulent Tubercle Bacilli on the Migration of Polymorphonuclear Leukocytes

STRAIN OF *M. tuberculosis*	CONCENTRATION OF BACTERIAL SUSPENSIONS	INHIBITORY EFFECT ON MIGRATION
H37Rv (virulent)	undiluted	++++
H37Rv (virulent)	1:30	++++
H37Rv (virulent)	1:90	+++
H37Rv (virulent)	1:270	+
H37Ra (avirulent)	undiluted	++
H37Ra (avirulent)	1:30	0

inhibition of migration results. This observation points to the possibility that the difference between the virulent and avirulent cultures of this strain might be quantitative rather than qualitative (2, 3)[4].

The experiments with charcoal and India ink show that indifferent particulate matter engulfed in large number by leukocytes do not inhibit their migration although a heavy charge of engulfed material might reduce or completely stop any further phagocytic activity (4).

In the present experiments, heat-killed virulent and avirulent tubercle bacilli had the same effect on migration as living ones. This action therefore seems to be caused by a cellular constituent of the virulent bacilli and does not directly depend on their multiplication.

The mechanism of the inhibitory action has not been elucidated by these experiments. It is possible that interference with locomotion can occur in the absence of other evidence of injury. The observation that large amounts of aviru-

[4] Although the H37Ra culture used did not produce a progressive disease in either guinea pigs or mice, it appeared to be a mixture of at least two variants, one of which formed clumps and the other formed weak but distinct serpentine cords in oleic acid-albumin medium. This suggests that the two variants may not be of identical virulence (Middlebrook, G., Dubos, R. J., and Pierce, C., J. Exper. Med., 1947, *86*, 175).

lent bacilli did not produce any detectable injury *in vivo* and yet can have some inhibitory action on locomotion *in vitro* points to this possibility.

SUMMARY

A method is described which is suitable for studying the migration of polymorphonuclear leukocytes *in vitro*. The migration of leukocytes which have engulfed virulent tubercle bacilli is completely inhibited, whereas a similar effect is not produced by avirulent bacilli taken up by polymorphonuclear leukocytes.

SUMARIO

El Efecto in Vitro de los Bacilos Tuberculosos sobre la Migración de los Fagocitos

La técnica descrita se presta para estudiar la migración de los leucocitos polimorfonucleares *in vitro*. La migración de los que han ingerido bacilos tuberculosos virulentos queda completamente inhibida, en tanto que no producen efecto semejante los bacilos avirulentos absorbidos por los leucocitos polimorfonucleares.

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VIRULENCE AND TUBERCULOGENIC STUDIES OF SIXTY CONSECUTIVE WEEKLY LOTS OF BCG VACCINE PRODUCED BY STANDARD TECHNIQUE[1,2]

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INTRODUCTION

World-wide acceptance and use of BCG emphasize the significance of methods that insure uniformly safe and potent vaccine preparations. Thus a problem of pressing importance is the development of a standard method for preparing a vaccine that will maintain a high level of viable organisms with stable tuberculogenic, allergenic, and immunizing properties. Of no less importance is the development of a quantitative method that permits periodic assessments of BCG's relative avirulence. Such a method is long overdue since only living vaccine is effective and the chance of a spontaneous enhancement of virulence is still thought of as a possibility by competent and unbiased scientists.

No uniform method for production of BCG vaccine is generally employed today. It is well known that many bacteriologic laboratories make radical alterations in Calmette's original technique (1 to 5). According to Jensen (4) the sole quantitative method which will detect variations in the "virulence" of BCG with some accuracy is a periodic intracutaneous guinea pig test using graded doses of from 0.1 to 0.0001 mg. of BCG vaccine. By these means Jensen was able to detect differences in "virulence" between a stock BCG strain which, contrary to Calmette's technique, had been maintained on Sauton medium from 1927 to 1931, and a new strain received from the Pasteur Institute in Paris in 1931. While the former strain produced frequent nodules with 0.001 mg. and large ones with 0.01 mg., the latter rarely produced nodules with 0.001 mg., and only small ones with 0.01 mg. If the term *virulent* is reserved to indicate organisms which give rise to a rapidly fatal infection or are able to reproduce increasingly in the tissues, it may be questioned whether Jensen's procedure is a proper test with which to determine the virulence of BCG. The reason for this is the fact that the skin lesions produced by these graded doses of microorganisms (dead tubercle bacilli will do the same) are observed for a period of only three weeks after inoculation. Jensen's procedure is an excellent quantitative test of BCG's tuberculogenic potency, the production of a localized and rapidly healing skin lesion, but is hardly a test of virulence.

When the preparation of vaccine was undertaken by the New York State Division of Laboratories and Research in 1946, plans were immediately made to evaluate methods for the preparation and biologic standardization of BCG. The present report describes the methods used and the results of assays made of each lot of vaccine prepared during a twelve month period.

[1] From the Division of Laboratories and Research, New York State Department of Health, Albany, New York.

[2] Presented before Le Premier Congrès International du BCG, Paris, France, June 18–23, 1948.

METHOD OF BCG VACCINE PRODUCTION

Potato cultures: The composition of Calmette's glycerolated bile-potato and water-potato media have not been altered. Two strains of BCG (No. 805. Series 1 and 2) were obtained from the Pasteur Institute in September, 1946. These strains were found to possess identical tuberculogenic, allergenic, and immunizing properties. They have since been transferred every two weeks in a locked laboratory and maintained in a locked incubator in which no other strain of *M. tuberculosis* or other bacteria are kept. Each set consists of one glycerolated-water and one glycerolated-bile potato subculture. The former is the seed from which the film on its liquid portion is transferred to Sauton medium for vaccine production. The glycerolated-bile potato subculture is for attenuation of virulence, according to Calmette's (6) contention, but more practically for restoration of the veil-like film production on glycerolated-water potato cultures which by six to eight successive biweekly passages has a tendency to grow roughly and less luxuriantly than following the first passages. For this purpose, Calmette's original technique has been followed, which consists of interposing one or two biweekly passages of BCG on the glycerolated-bile potato medium every two months. Since film development on the liquid portion of the glycerolated-water potato medium is essential for the seeding of Sauton medium for subsequent vaccine production, it is the practice at present to tilt the one week old glycerolated-water potato culture on the side until the liquid frees the small fragments of the veil-like film along the edges of the potato surface and floats them onto the liquid portion of the medium. This insures a luxuriant supply of film for transfers to Sauton medium the following week.

Sauton cultures: The composition of Sauton medium has not been altered. Ehrlenmeyer flasks (350 ml.) containing 160 ml. of Sauton medium have been used exclusively. The veil-like film in the two week old glycerolated-water potato culture is transferred with a 3 by 1 cm. platinum spatula to the surface of the Sauton medium. At 37.5°C. the film covers about half the surface in three days and the entire surface as well as a few cm. on the sides of the flask in seven days. Such growth within one week is used as the criterion of a normal BCG Sauton I culture. At this time transfers are made with a spiral-shaped platinum wire spade, having a diameter of 25 mm. The film along the border is carefully scooped onto the spade in folded fashion and this is again floated on Sauton medium where it flattens out promptly. One spadeful is sufficient for each Sauton II flask. These cultures grow much more rapidly than Sauton I cultures and the entire surface is usually covered with film in four days. After ten days' growth at 37.5°C., the densely coiled and 3 to 5 mm. thick amber-colored film is harvested for vaccine. This is an alteration in Calmette's original technique which specifies 20 to 25 day old Sauton II culture for vaccine production. The custom in many laboratories is to use 21 to 30 day old Sauton II cultures (2, 7). It was decided to shorten the incubation of the Sauton II cultures to 10 to 11 days when serial Loewenstein cultures revealed that such cultures contained between 20 and 40 million living bacilli per mg. of semidry weight or 80 to 160 million living bacilli per mg. of absolutely dry vaccine while the 3 to 4 week old Sauton II cultures contained approximately one-half that number. The results of vaccination with the 10 to 11 day old Sauton II culture vaccine have been uniformly better, especially with the multiple puncture technique, than with vaccine prepared from the 21 to 30 day old Sauton II cultures.

Semidry and dry weight of BCG in 10 to 11 day old Sauton II Cultures: For the purpose of determining the growth curve of BCG in 10 to 11 day old Sauton II cultures in 160 ml. of medium it has been the practice to single out such a culture once a week for one year for exact determination of the semidry and dry weight. The culture is emptied into a Büchner suction filter supplied with a fine filter paper (Whatman No. 3) which retains the smallest particles of BCG. Suction is continued until no more liquid is expressed from the mass. After the semidry weight is determined, the mass is placed in a dry sterilizer at 100°C. until a constant weight is obtained. By grouping these cultures together into twelve weekly lots for the purpose of statistical analysis, the data presented in table 1 and summarized in figure 2 were obtained.

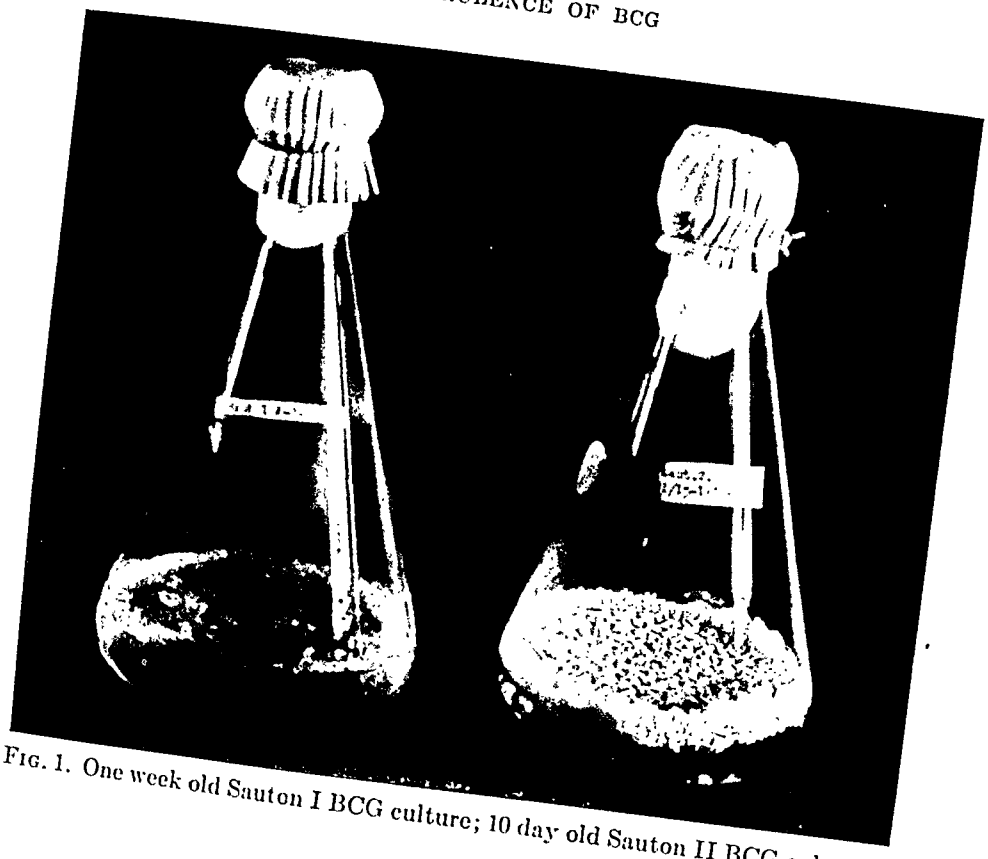

FIG. 1. One week old Sauton I BCG culture; 10 day old Sauton II BCG culture.

TABLE 1

Weight of BCG in Sauton II Cultures

Statistical analysis of semidry and dry weight (Gm.) of BCG harvested from 160 ml. Sauton medium after 10 to 11 days' incubation at 37.5°C.

(Twelve cultures in each group)

WEEKLY LOTS OF SAUTON II CULTURES	SEMIDRY WEIGHT		DRY WEIGHT		RATIO
	Average	Sigma	Average	Sigma	
1 to 12 weeks	5.971	0.503	1.475	0.089	4.05:1
13 to 24 weeks	5.733	0.421	1.456	0.088	3.94:1
25 to 36 weeks	5.675	0.350	1.480	0.100	3.83:1
37 to 48 weeks	5.669	0.495	1.428	0.084	3.97:1
49 to 60 weeks	5.715	0.339	1.452	0.128	3.94:1

WEEKLY GROUP COMPARISONS	Probability*			
	t	P	t	P
1 to 12 vs. 13 to 24	1.200	0.23	0.134	0.88
1 to 12 vs. 25 to 36	1.602	0.14	0.034	0.90
1 to 12 vs. 37 to 48	1.418	0.20	1.278	0.23
1 to 12 vs. 49 to 60	1.398	0.21	0.654	0.32

* Significant differences ($P \leqq 0.01$) require that $t \geqq 2.819$ when each group contains 12 samples (8).

It will be noted that a remarkable stability obtained in the semidry and dry weights throughout the entire 60 week observation period and that not a single significant deviation occurred from the first twelve weekly lots of Sauton II cultures to the final lots (49 to 60 weeks). The ratio between the semidry and dry BCG is almost constantly 4:1. Thus the 20 mg. per ml. BCG vaccine used for transcutaneous vaccination methods actually contains 5 mg. of dry BCG per ml., and the 1.0 mg. per ml. BCG vaccine for intracutaneous inoculation contains 0.25 mg. dry BCG per ml.

An attempt was also made to determine the exact population of living microorganisms per milligram of BCG in every weekly lot of vaccine distributed. This is accomplished by serial dilutions of exactly weighed vaccine and by seeding 1 ml. of each of the 10^{-5}, 10^{-6}, 10^{-7}, and 10^{-8} dilutions on 5 large tubes of Loewenstein's egg medium. This study is still incomplete.

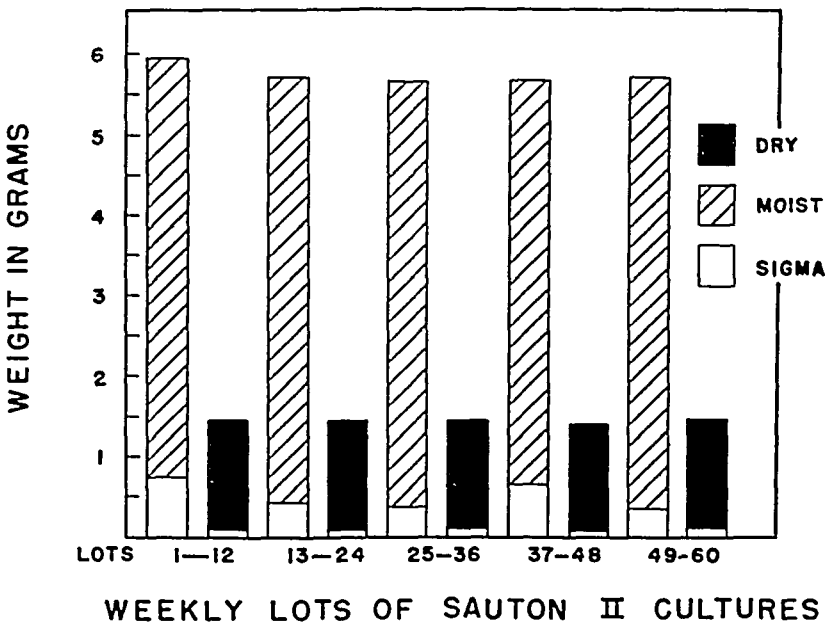

FIG. 2. Average semidry and dry weights of sixty consecutive weekly lots of vaccine produced in 160 ml. Sauton 10 to 11 day old cultures of BCG.

Preparation of vaccine: The traditional Calmette method of harvesting the Sauton II cultures for preparation of vaccine dispenses with the actual weighing of the BCG mass, on the assumption that the 21 to 30 day old 150 ml. Sauton culture contains 5 Gm. semidry BCG. This rule is not absolute and many laboratories desiring a more or less constant concentration of vaccine resort to transferring BCG aseptically to filter paper between which the excess liquid is pressed out. The semidry mass is then weighed in sterile porcelain crucibles after which it is emptied into a flask containing glass or steel balls. In a ball mill the vaccine is mixed with dilution fluid to the proper concentration. To reduce contamination, a harvesting apparatus (figure 3) has been constructed which combines all these operations in a closed system. The slightly agitated Sauton II culture is emptied into a funnel which leads to a cylinder at the bottom of which a strainer filters out the culture. A piston is pressed against the BCG mass within the cylinder until no more liquid is expressed. The shaft of the piston is calibrated and indicates the height of the column of BCG within the cylinder. A nomogram converts height into grams of semidry BCG. The strainer is pushed aside and a quick tap on the handle of the piston releases the column of BCG which drops into a glass cylinder supplied with 1 Kg. of 4 mm. wide and 0.5 Kg. of 3 mm. wide SKF inoxidizable steel balls. Before adding diluent, the ball mill is rotated at 30 r.p.m. for two

to three minutes until the BCG mass has become finely divided into an oily paste and adheres to the sides of the glass cylinder and the steel balls.

The diluent recommended by Calmette is composed of one part of Sauton medium and three parts of distilled water. In order to make the vaccine isotonic and more easily tolerated when injected intracutaneously, physiologic salt solution has been substituted for the distilled water. When the mass of BCG has been sufficiently homogenized, about 10 ml. of diluent are added and the ball mill is rotated for two minutes until a semiliquid paste is formed. Sufficient diluent is then added to make the final concentration of 20 mg. per

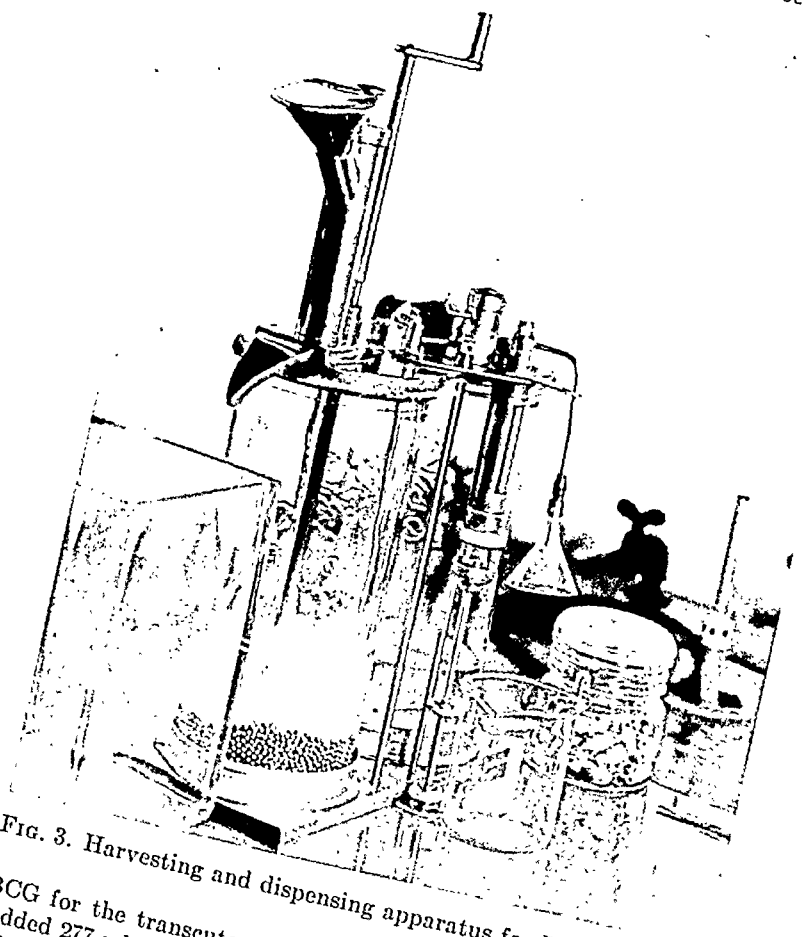

FIG. 3. Harvesting and dispensing apparatus for BCG vaccine.

ml. semidry BCG for the transcutaneous methods of vaccination, *i.e.*, to 5.540 Gm. of BCG mass is added 277 ml. of diluent. The ball mill is now rotated at 30 r.p.m. for five minutes. To the glass cylinder on the right in figure 4 is added 5 ml. of the 20 mg. per ml. BCG vaccine (100 mg.) per 95 ml. of diluent in order to make 1.0 mg. per ml. BCG for the intracutaneous method of vaccination. As mentioned before, these concentrations contain 5.0 mg. per ml. and 0.25 mg. per ml. dry BCG respectively.

As shown in figure 4, the vaccine from either glass cylinder is dispensed in 5 and 10 ml. vials by means of a one way suction system that is operated by a foot pedal. By means of this apparatus, one person may fill, stopper, and seal 8 to 10 vials per minute.

Tests for purity of the vaccine: Control tests to rule out the presence of contaminating microorganisms in the vaccine are made by inoculating 0.5 ml. of the fluid portion of the

Sauton II culture, before the film is harvested, into 20 ml. of meat-infusion semisolid medium (*FS2B*) (9); 20 ml. of meat-infusion broth, aerobic and anaerobic (*FS3B*); and in a pour plate using beef-infusion agar with 0.2 per cent of glucose (*FS5A*). These tests are repeated with the 20 mg. per ml. and the 1.0 mg. per ml. vaccines in bulk before dispensing, with the residue after dispensing, and with one vial of each twenty dispensed. A duplicate flask of 100 ml. of the 1.0 mg. per ml. material in bulk is tested simultaneously by a member of the sterility test group of the Antitoxin, Serum, and Vaccine Laboratories. A volume of 160 ml. of meat-infusion semisolid medium (*FS2B*) is inoculated with 5 to 5.5 ml. of the material; two aerobic and two anaerobic tubes of meat-infusion broth (*FS3B*) with 0.5 ml. each.

If no growth of contaminating microorganisms appears in the various inoculated media after twenty-four hours' incubation, the vaccine is distributed for a period of seven days.

FIG. 4. Dispensing BCG vaccine in vials.

Preliminary studies indicate that the efficacy of the vaccine is not impaired by the addition to the diluent of 20 units of penicillin per ml. of the intracutaneous vaccine. This concentration of penicillin completely inhibits the growth of staphylococci and streptococci during the 10 day period recommended for use of the vaccine before it is returned to the Division of Laboratories and Research. This precaution is taken only with the 1.0 mg. per ml. vaccine because penicillin is completely destroyed by the 20 mg. per ml. vaccine within a few days.

QUANTITATIVE TESTS FOR STABILITY OF BCG'S VIRULENCE

For every lot of BCG vaccine prepared weekly, two tuberculin-negative guinea pigs weighing approximately 350 Gm. are injected subcutaneously in the left hind leg with 0.5 ml. of the 20 mg. per ml. vaccine, or 10 mg. semidry weight of BCG. Tuberculin tests are repeated at monthly intervals. One month after inoculation, 100 per cent positive edematous reactions, which exceed 10 by 10 mm. in

diameter within forty-eight hours, are obtained with 10 mg. of Old Tuberculin. Two months after inoculation, approximately 85 per cent positive edematous reactions are obtained with 0.01 mg. of Old Tuberculin. Six months after inoculation, positive reactions occur in approximately 70 per cent of the animals tested with 0.001 mg. of Old Tuberculin. Throughout the entire six month period, 0.1 mg. Old Tuberculin yields positive reactions in 100 per cent of the animals tested.

Skin puncture through a drop of either living or dead BCG (20 mg. per ml.) results in an accelerated tubercle formation, consisting of erythema, induration and suppuration. This reaction is observed regularly in these animals from three weeks after inoculation until they are killed three and six months after inoculation. The accelerated reaction begins a few hours after the puncture is made, reaches its height in six to ten days, and heals completely within a month. In the normal guinea pig such a puncture slowly leads to a papular development in one week, reaching its height in three weeks when it subsides slowly without any tendency to suppuration. After two months no trace is found of the original lesion, while in the allergic animals a slightly pigmented and sunken area in the skin invariably remains.

The only visible manifestation of the subcutaneous inoculation with BCG is a rapidly developing sterile abscess in the left hind leg. Two weeks after inoculation the leg becomes markedly swollen and the regional lymph nodes are palpably enlarged. One month after inoculation the tumefaction is larger and fluctuation is present. The regional lymph nodes increase in size but remain firm in consistency. Thenceforth the abscess becomes stationary and occasionally breaks down with the discharge of a semisolid caseum rich in acid-fast bacilli. The ruptured abscesses heal promptly but the regional lymph nodes remain hypertrophied and hard.

The weight of animals reflects their apparently normal condition except for the left hind leg abscess and regional lymphadenitis. Table 2 presents the average weights of ten groups, each consisting of 12 inoculated animals. In addition, the weights are recorded of four groups when they were killed three months after inoculation and three groups which were killed six months after inoculation.

It will be noted that none of the groups differed significantly from each other before inoculation and that the same increments in weight had taken place in three and six months, respectively, without any significant deviations between any of the groups. It is apparent, therefore, that the low grade infection with BCG interfered but little with the normal growth of these animals. This fact is confirmed by the 1940 experience (10) in which the growth curve of 24 normal guinea pigs weighing 297 sigma 53.8 Gm. was plotted for 53 preinfection days and 102 days after the intraperitoneal inoculation of 0.0001 mg. (302 viable) human tubercle bacilli. At the end of that period the average body weight at autopsy was 374 sigma 67.3 Gm.

Quantitative assessment of autopsy material: One of each weekly pair of guinea pigs, which had been inoculated subcutaneously in the left hind leg with 10 mg. of BCG, was killed at three and the other at six months after the inoculation.

Autopsy was delayed until the carcass had remained in the refrigerator (2° to 4°C.) for several hours in order to allow the blood to coagulate. This insures uniformity in the quantitative assessment of the volume and weight of various

TABLE 2

Weight of Animals

Statistical analysis of weight (Gm.) of guinea pigs before inoculation with 10 mg. BCG and when killed 3 months (Series A) and 6 months (Series B) afterwards

(Twelve animals in each group)

GROUPS OF ANIMALS	BEFORE INOCULATION			
	Series A		Series B	
	Average	Sigma	Average	Sigma
weeks				
1 to 12	355	28.51	360	29.11
13 to 24	353	33.44	349	40.38
25 to 36	331	37.53	334	39.60
37 to 48	383	58.12	377	38.82
49 to 60	336	42.17	321	53.62
Probability				
GROUP COMPARISONS	*t*	P	*t*	P
1 to 12 vs. 13 to 24	0.220	0.82	0.750	0.45
1 to 12 vs. 25 to 36	1.729	0.09	0.686	0.50
1 to 12 vs. 37 to 48	1.446	0.18	0.701	0.48
1 to 12 vs. 49 to 60	0.186	0.85	2.131	0.04

GROUPS OF ANIMALS	3 MONTHS AFTERWARDS		6 MONTHS AFTERWARDS	
	Average	Sigma	Average	Sigma
weeks				
1 to 12	616	68.33	743	63.52
13 to 24	634	69.61	764	62.27
25 to 36	656	53.77	791	84.90
37 to 48	656	83.45	—	—
Probability				
GROUP COMPARISONS	*t*	P	*t*	P
1 to 12 vs. 13 to 24	0.588	0.58	0.784	0.46
1 to 12 vs. 25 to 36	1.470	0.15	1.494	0.15
1 to 12 vs. 37 to 48	1.225	0.25	—	—

lymph nodes and organs. The major lymph nodes mentioned in tables 3 and 4 were carefully dissected free from contiguous tissue and their volume determined by water displacement in a specially constructed apparatus (figure 5) with cylinders calibrated to 0.001 ml. The volume of the spleen, liver, and lungs

was determined in the same manner. Finally, the weight of these organs and the pooled lymph nodes was expressed in percentage of the animal's body weight.

TABLE 3

Volume of Lymph Nodes

Statistical analysis of volume (ml.) of lymph nodes in guinea pigs killed 3 months after subcutaneous inoculation in left hind leg with 10 mg. BCG

(Twelve animals in each group)

LYMPH NODES	INOCULATED WITH WEEKLY LOTS OF BCG VACCINE							
	1 to 12 weeks (1)		13 to 24 weeks (2)		25 to 36 weeks (3)		37 to 48 weeks (4)	
	Average	Sigma	Average	Sigma	Average	Sigma	Average	Sigma
Lf. sup. inguinal	0.148	0.071	0.146	0.087	0.157	0.027	0.145	0.091
Rt. sup. inguinal	0.056	0.024	0.051	0.030	0.060	0.024	0.059	0.029
Lf. deep inguinal	0.018	0.011	0.018	0.013	0.018	0.011	0.017	0.012
Rt. deep inguinal	0.007	0.004	0.011	0.006	0.009	0.006	0.008	0.004
Lf. femoral	0.111	0.034	0.110	0.049	0.117	0.053	0.117	0.045
Rt. femoral	0.030	0.028	0.035	0.017	0.028	0.027	0.024	0.017
Periportal	0.010	0.009	0.012	0.014	0.008	0.004	0.007	0.006
Lf. tracheobronchial	0.096	0.055	0.099	0.027	0.093	0.048	0.094	0.062
Rt. tracheobronchial	0.079	0.025	0.078	0.029	0.084	0.038	0.084	0.031
Lf. cervical	0.081	0.014	0.073	0.024	0.080	0.020	0.079	0.021
Rt. cervical	0.079	0.012	0.072	0.017	0.079	0.025	0.082	0.015
Lf. axillary	0.064	0.015	0.066	0.019	0.066	0.014	0.062	0.018
Rt. axillary	0.066	0.022	0.061	0.019	0.066	0.023	0.058	0.018

Probability

LYMPH NODES	(1) vs. (2)		(1) vs. (3)		(1) vs. (4)	
	t	P	*t*	P	*t*	P
Lf. sup. inguinal	0.360	0.68	0.270	0.78	0.086	0.90
Rt. sup. inguinal	0.255	0.81	1.960	0.07	0.262	0.79
Lf. deep inguinal	0.032	0.90	0.027	0.90	0.020	0.90
Rt. deep inguinal	1.960	0.07	0.980	0.38	0.409	0.66
Lf. femoral	0.025	0.90	0.318	0.75	0.357	0.72
Rt. femoral	0.509	0.62	0.173	0.88	0.612	0.32
Periportal	0.377	0.72	0.701	0.49	0.919	0.38
Lf. tracheobronchial	0.016	0.90	0.014	0.89	0.007	0.90
Rt. tracheobronchial	0.009	0.90	0.360	0.72	0.421	0.68
Lf. cervical	1.298	0.23	0.044	0.90	0.046	0.90
Rt. cervical	1.641	0.14	0.049	0.90	0.034	0.90
Lf. axillary	1.347	0.21	1.641	0.14	0.044	0.90
Rt. axillary	0.539	0.59	0.145	0.89	0.051	0.90

By this procedure an attempt was made to assess the degree of hypertrophy caused by the 10 mg. BCG inoculum.

Volume of lymph nodes: The volumetric data on thirteen different lymph

nodes removed from the four groups of animals which were killed three months after inoculation are presented in table 3 and figure 6, and the data for the three

TABLE 4

Volume of Lymph Nodes

Statistical analysis of volume (ml.) of lymph nodes in guinea pigs killed 6 months after subcutaneous inoculation in left hind leg with 10 mg. BCG

(Twelve animals in each group)

LYMPH NODES	INOCULATED WITH WEEKLY LOTS OF BCG VACCINE					
	1 to 12 weeks (1)		13 to 24 weeks (2)		25 to 36 weeks (3)	
	Average	Sigma	Average	Sigma	Average	Sigma
Lf. sup. inguinal	0.155	0.064	0.150	0.069	0.179	0.089
Rt. sup. inguinal	0.051	0.026	0.061	0.029	0.062	0.023
Lf. deep inguinal	0.022	0.014	0.023	0.019	0.025	0.017
Rt. deep inguinal	0.008	0.004	0.010	0.007	0.008	0.004
Lf. femoral	0.105	0.052	0.105	0.048	0.115	0.045
Rt. femoral	0.032	0.014	0.039	0.017	0.040	0.015
Periportal	0.008	0.006	0.010	0.009	0.007	0.002
Lf. tracheobronchial	0.096	0.035	0.093	0.035	0.091	0.060
Rt. tracheobronchial	0.077	0.042	0.086	0.042	0.089	0.039
Lf. cervical	0.073	0.019	0.066	0.016	0.074	0.017
Rt. cervical	0.069	0.018	0.066	0.016	0.076	0.017
Lf. axillary	0.059	0.021	0.061	0.017	0.061	0.010
Rt. axillary	0.064	0.024	0.061	0.021	0.060	0.015

Probability

LYMPH NODES	(1) vs. (2)		(1) vs. (3)	
	t	P	*t*	P
Lf. sup. inguinal	0.145	0.87	0.265	0.79
Rt. sup. inguinal	0.845	0.41	1.036	0.31
Lf. deep inguinal	0.137	0.88	0.270	0.78
Rt. deep inguinal	0.543	0.59	0.019	0.90
Lf. femoral	0.129	0.90	0.480	0.65
Rt. femoral	1.009	0.32	1.398	0.14
Periportal	0.613	0.52	0.980	0.32
Lf. tracheobronchial	0.016	0.90	0.024	0.90
Rt. tracheobronchial	0.502	0.65	0.701	0.49
Lf. cervical	0.044	0.90	0.046	0.90
Rt. cervical	0.042	0.90	1.347	0.21
Lf. axillary	0.049	0.90	0.042	0.90
Rt. axillary	0.058	0.90	0.051	0.90

groups of animals killed six months after inoculation are presented in table 4 and figure 7. It will be noted that the six sets of nodes situated on the left side of the body (inoculated side) are symmetrical with the six sets of nodes on the right

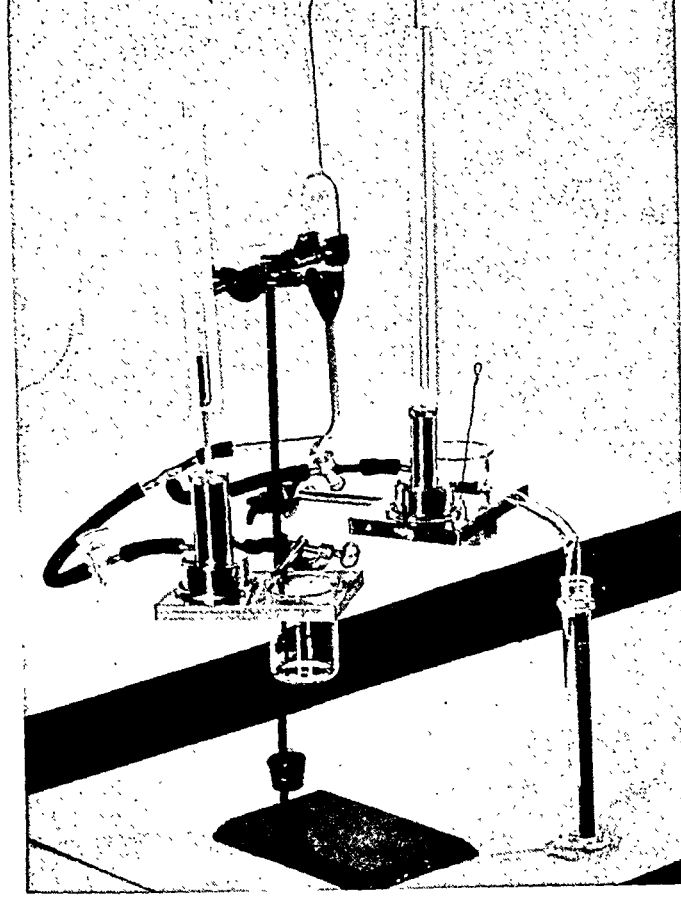

FIG. 5. Volumetric apparatus for measurement of larger organs (left cylinder) and smaller nodes (right cylinder).

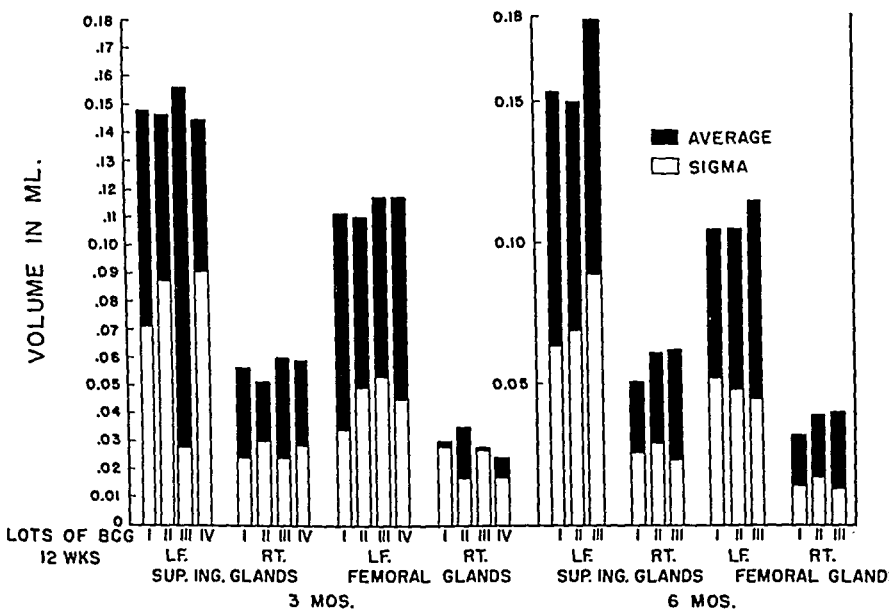

FIG. 6. Average volume of left and right superficial inguinal and femoral lymph nodes in guinea pigs killed three or six months after subcutaneous injection in left hind leg with 10 mg. of sixty consecutive weekly lots of BCG vaccine.

side of the body. Thus the only unpaired lymph node is the periportal node. The measurements of the lymph nodes on the right side may therefore be considered as controls for the lymph nodes on the left side. Thus in tables 3 and 4 it may be seen that the left side superficial and deep inguinal and femoral nodes, which are directly draining the BCG abscess in the left hind leg, are regularly about three times larger in size than the corresponding nodes on the right side of the body. The remaining nodes show no appreciable differences between the left and right sides of the body. It will also be noted in tables 3 and 4 that the hypertrophy of the left superficial and deep inguinal and the femoral nodes pre-

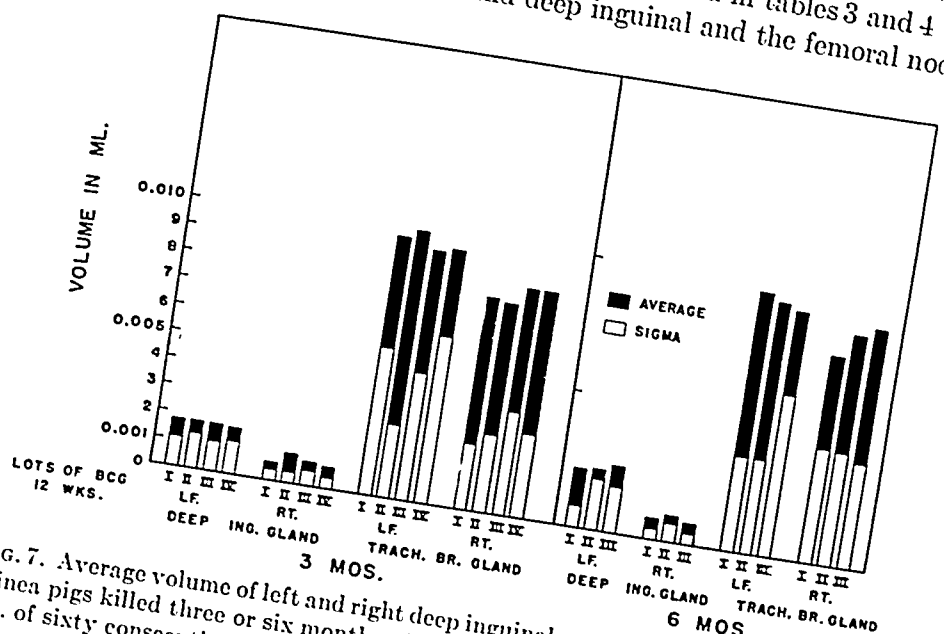

FIG. 7. Average volume of left and right deep inguinal and tracheobronchial lymph nodes in guinea pigs killed three or six months after subcutaneous injection in left hind leg with 10 mg. of sixty consecutive weekly lots of BCG vaccine.

sent no marked increments in size from three to six months after the animals were inoculated with BCG. The frequency of caseum in these nodes was greater three months after inoculation than after six months. Calcified nodes were observed in the reverse order. The frequency of acid-fast bacilli in smears and of colonies in cultures made with caseous material from left superficial and deep inguinal and femoral nodes was greater after three than after six months' inoculation.

The impact on the guinea pig's lymphatic system by a subcutaneous inoculation with 0.1 mg. of virulent human tubercle bacilli (approximately 32,000 viable microorganisms) in the left hind leg, the identical route used with BCG inoculation, is demonstrated by the 1940 experience (11) with 24 control guinea pigs which died from generalized tuberculosis 47 to 51 days later. The volume of the lymph nodes at necropsy was as follows:

LYMPH NODES	VOLUME IN ML.	
	Average	Sigma
Left superficial inguinal	0.570	0.319
Left deep inguinal	0.213	0.217
Left femoral	0.359	0.169
Periportal	0.708	0.352
Left tracheobronchial	0.528	0.265
Right tracheobronchial	0.413	0.209
Left cervical	0.339	0.163
Right cervical	0.323	0.153
Left axillary	0.295	0.191
Right axillary	0.243	0.129

TABLE 5

Weight of Spleen, Liver, Lungs, and Lymph Nodes
Statistical analysis of weight in per cent of body weight of the spleen, liver, lungs, and lymph nodes in guinea pigs killed 3 months after subcutaneous inoculation with 10 mg. BCG

(Twelve animals in each group)

ORGANS	WEEKLY LOTS OF BCG VACCINE							
	1 to 12 weeks (1)		13 to 24 weeks (2)		25 to 36 weeks (3)		37 to 48 weeks (4)	
	Average	Sigma	Average	Sigma	Average	Sigma	Average	Sigma
Spleen	0.101	0.014	0.115	0.020	0.114	0.016	0.122	0.028
Liver	4.708	0.752	4.180	0.595	4.367	0.758	4.639	1.015
Lungs	1.119	0.273	1.079	0.229	0.898	0.176	1.088	0.188
Lymph nodes	0.165	0.045	0.171	0.047	0.160	0.036	0.161	0.062

Probability

ORGANS	(1) vs. (2)		(1) vs. (3)		(1) vs. (4)	
	t	P	*t*	P	*t*	P
Spleen	1.906	0.06	1.991	0.06	2.023	0.05
Liver	1.436	0.12	1.058	0.31	0.181	0.85
Lungs	0.372	0.72	2.254	0.04	0.221	0.82
Lymph nodes	0.255	0.81	0.254	0.81	0.172	0.88

Thus it will be noted that the virulent infection produced a generalized lymph node hypertrophy in contrast with the localized process in animals inoculated with 100 times greater dose of BCG, or approximately 400 million viable BCG.

Weight of the spleen, liver, lungs, and lymph nodes: The more exact quantitative data are those expressed in percentage of the body weight of the animal from which the organs were removed since the body weight varies from animal to animal. Table 5 presents such data for the spleen, liver, lungs, and lymph nodes

taken from four groups of animals killed three months after inoculation with BCG. Table 6 shows the data for animals killed six months after inoculation. The results are summarized in figure 8. For comparison, similar data from the above mentioned (11) virulently infected animals are as follows:

ORGANS	PER CENT OF BODY WEIGHT	
	Average	Sigma
Spleen	1.71	1.30
Liver	9.11	1.51
Lungs	3.25	0.81
Total lymph nodes	3.61	0.95

TABLE 6

Weight of Spleen, Liver, Lungs, and Lymph Nodes

Statistical analysis of weight in per cent of body weight of the spleen, liver, lungs, and lymph nodes in guinea pigs killed 6 months after subcutaneous inoculation with 10 mg. BCG

(Twelve animals in each group)

ORGANS	WEEKLY LOTS OF BCG VACCINE					
	1 to 12 weeks (1)		13 to 24 weeks (2)		25 to 36 weeks (3)	
	Average	Sigma	Average	Sigma	Average	Sigma
Spleen	0.099	0.029	0.095	0.015	0.109	0.030
Liver	4.023	0.545	4.216	0.971	4.493	0.607
Lungs	0.846	0.192	0.901	0.226	1.007	0.229
Lymph nodes	0.136	0.040	0.137	0.030	0.164	0.026

ORGANS	Probability			
	(1) vs. (2)		(1) vs. (3)	
	t	P	t	P
Spleen	0.409	0.65	0.791	0.42
Liver	0.573	0.58	1.568	0.14
Lungs	0.612	0.54	1.499	0.15
Lymph nodes	0.066	0.90	1.960	0.06

Table 5 shows that the weight of the spleen in the animals killed three months after inoculation with BCG varied between 0.101 and 0.122 per cent of the animals' body weight. The weight varied between 0.095 and 0.109 per cent of the animals' body weight at six months after inoculation (table 6). At no time was any significant deviation observed between any two groups in these series. Of greater importance is the fact that no macroscopic tubercle was ever found in the spleen of these animals.

The weight of the liver (tables 5 and 6) varied between 4.023 and 4.708 per cent of the animals' body weight three and six months after inoculation; the

lungs between 0.898 and 1.119 per cent of the animals' body weight; and finally, the total lymph nodes between 0.136 and 0.171 per cent. With the exceptions of the fibrocaseous hypertrophy observed in the left superficial and deep left inguinal nodes and the left femoral nodes adjacent to the BCG inoculation abscess, no macroscopic tuberculous lesions were seen in the liver and lungs of the animals killed three and six months respectively after inoculation with BCG from 48 different lots of vaccine prepared during one year.

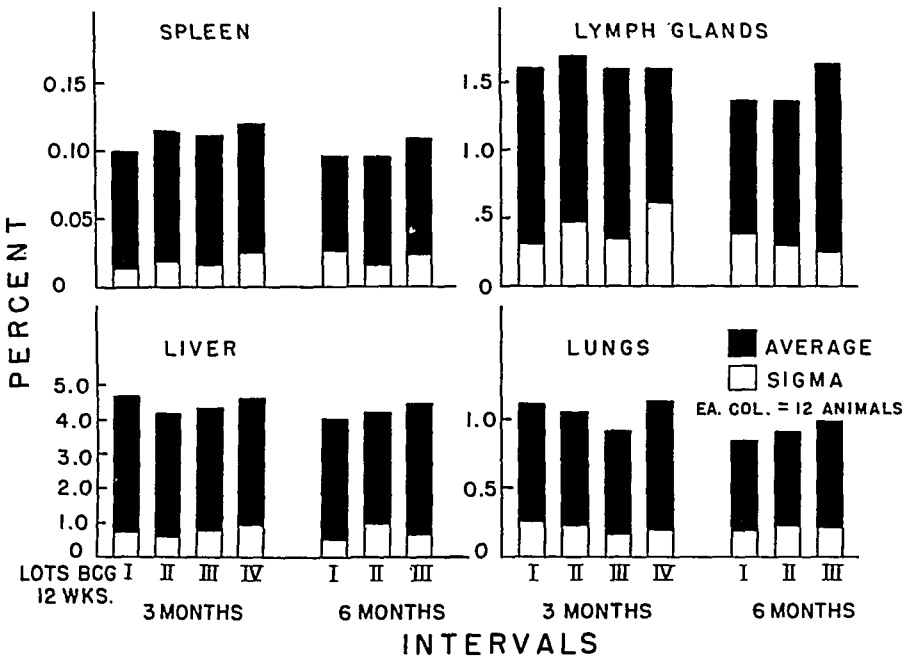

FIG. 8. Average weight of spleen, liver, lungs, and total lymph nodes in guinea pigs killed three or six months after subcutaneous injection in left hind leg with 10 mg. of sixty consecutive weekly lots of BCG vaccine.

Comment

The data on the lymph nodes, spleen, liver, and lungs indicate that the avirulence of BCG is not absolute and that the very low grade virulence of BCG is of a significantly stationary type for vaccine prepared by the method set forth in this presentation. There can be no question about the reliability of this quantitative procedure to detect significant deviations in BCG's virulence should such ever take place. It would seem desirable, therefore, that some such quantitative assessment of lesions produced in guinea pigs by each lot of vaccine prepared for use in man should become a standard procedure in order that laboratories entrusted with the production of vaccine at all times may be prepared to report objectively on the avirulence of their preparations.

BCG'S TUBERCULOGENIC POTENCY IN ANIMAL SKIN TESTS

Following Jensen's (4) study on variations in the "virulence" of different strains of BCG cultivated by different methods, it was decided from the very

start of the present investigation to evaluate the tuberculogenic potency of every lot of vaccine for one year. In an attempt to increase the objectivity of the investigation as much as possible, most of the observed data were reduced to measurements of the local skin lesion at one and three week intervals after inoculation. For this purpose a white tuberculin-negative guinea pig, weighing between 300 and 350 Gm. was used. The skin on the abdomen and the left side of the body was depilated and thereafter washed with alcohol and ether. Intracutaneous injections of 0.1 ml. of saline dilutions of the BCG vaccine containing 1.0 mg., 0.1 mg., 0.01 mg., and 0.001 mg. of vaccine (final 0.1, 0.01,

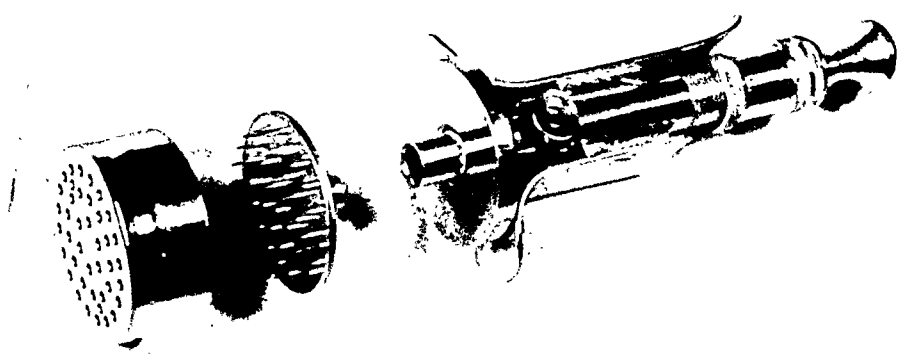

FIG. 9. Spring-actuated multiple puncture apparatus with detachable needle holder and head plate for individual BCG vaccination.

0.001, and 0.0001 mg.) were made in the four quadrants of the abdomen. Care was taken to inject the vaccine as superficially as possible so that a white bleb remained visible for about five minutes. Three to five drops of the transcutaneous (20 mg. per ml.) vaccine were spread evenly across the depilated skin on the left side of the body. Holding the skin taut, forty punctures, approximately 3 mm. in depth, were made through the skin with the automatic multiple apparatus (12) (figure 9). The skin was stretched slightly in order to verify the presence of petechial bleeding from each puncture. One or two drops of the vaccine were then spread across the punctured area and the animal replaced in its cage.

Typical vaccination nodules with suppurating centers were produced regularly with 0.1 and 0.01 mg. of BCG vaccine and only a hardened nodule with 0.001

mg. No lesion was produced by 0.0001 mg. of the vaccine. After one week, the multiple puncture sites presented approximately 1.5 mm. wide red, raised, and firm papules. These measured approximately 2.6 mm. in width and 1.5 mm. in height after three weeks. Desquamation and healing occurred after four weeks and were complete three to four weeks later. Suppuration in individual papules was exceedingly rare. For statistical analysis, readings of the intracutaneous and transcutaneous lesions were most advantageously made one and three weeks after inoculation. At the latter time the maximum development was reached. Tables 7 and 8 present the data on 60 individual guinea

TABLE 7

BCG Skin Lesions

Statistical analysis of maximum diameter (mm.) of skin lesions produced in guinea pigs one week after intracutaneous and transcutaneous inoculations with graded doses of BCG

(Twelve animals in each group)

WEEKLY LOTS OF BCG VACCINE	INTRACUTANEOUS INJECTION						MULTIPLE PUNCTURE	
	0.1 mg.		0.01 mg.		0.001 mg.		20 mg./ml.	
	Average	Sigma	Average	Sigma	Average	Sigma	Average	Sigma
1 to 12 weeks	5.75	0.59	4.33	0.74	2.33	0.62	1.63	0.22
13 to 24 weeks	5.42	0.49	3.83	0.79	2.25	0.43	1.54	0.13
25 to 36 weeks	5.67	0.65	4.09	0.64	2.33	0.62	1.54	0.28
37 to 48 weeks	6.13	0.49	4.33	0.62	2.75	0.43	1.75	0.33
49 to 60 weeks	6.50	0.50	4.59	0.76	2.67	0.47	1.88	0.69

COMPARISON OF WEEKLY GROUPS	Probability							
	t	P	*t*	P	*t*	P	*t*	P
1 to 12 vs. 13 to 24	1.416	0.18	1.512	0.15	0.754	0.48	1.161	0.28
1 to 12 vs. 25 to 36	0.029	0.90	0.806	0.41	1.592	0.15	0.848	0.41
1 to 12 vs. 37 to 48	1.634	0.12	1.764	0.11	1.838	0.08	1.014	0.29
1 to 12 vs. 49 to 60	0.316	0.78	0.931	0.32	1.470	0.18	2.663	0.02

pigs, which for statistical purposes have been subdivided into groups of 12 animals, each of which was inoculated with successive lots of vaccine prepared at weekly intervals during sixty weeks. These data are summarized in figure 10.

Comment

One is struck by the relative stability of the skin reactions produced by each graded dose of BCG vaccine both after one week and three weeks, whether tested by the intracutaneous or transcutaneous routes. In table 7 it would appear that the 0.1 mg. dose of BCG grew more tuberculogenic from the first one to 12 weekly lots, which produced a 5.75 sigma 0.59 mm. wide suppurating infiltration in the skin, until the final 49 to 60 weekly lots which produced 6.50 sigma 0.50 mm. similar lesions. The multiple puncture lesions produced by the same lots of

TABLE 8

BCG Skin Lesions

Statistical analysis of maximum diameter (mm.) of skin lesions produced in guinea pigs three weeks after intracutaneous and transcutaneous inoculations with graded doses of BCG

(Twelve animals in each group)

WEEKLY LOTS OF BCG VACCINE	INTRACUTANEOUS INJECTION						MULTIPLE PUNCTURE	
	0.1 mg.		0.01 mg.		0.001 mg.		20 mg./ml.	
	Average	Sigma	Average	Sigma	Average	Sigma	Average	Sigma
1 to 12 weeks	6.92	0.76	5.17	0.80	3.42	0.45	2.75	0.25
13 to 24 weeks	7.09	0.82	5.25	0.60	3.09	0.42	2.38	0.46
25 to 36 weeks	6.92	0.64	5.75	0.60	3.42	0.47	2.42	0.34
37 to 48 weeks	7.25	0.44	5.67	0.49	3.67	0.74	2.59	0.34
49 to 60 weeks	7.17	0.38	5.92	0.36	3.59	0.39	2.79	0.38

COMPARISON OF WEEKLY GROUPS	Probability							
	t	P	*t*	P	*t*	P	*t*	P
1 to 12 vs. 13 to 24	0.502	0.61	0.264	0.79	1.795	0.09	2.322	0.04
1 to 12 vs. 25 to 36	1.568	0.15	1.920	0.07	1.176	0.28	0.760	0.48
1 to 12 vs. 37 to 48	1.245	0.23	1.749	0.09	0.943	0.32	1.223	0.22
1 to 12 vs. 49 to 60	0.973	0.35	*2.824*	*0.01*	0.946	0.37	0.279	0.79

Significant deviation italicized.

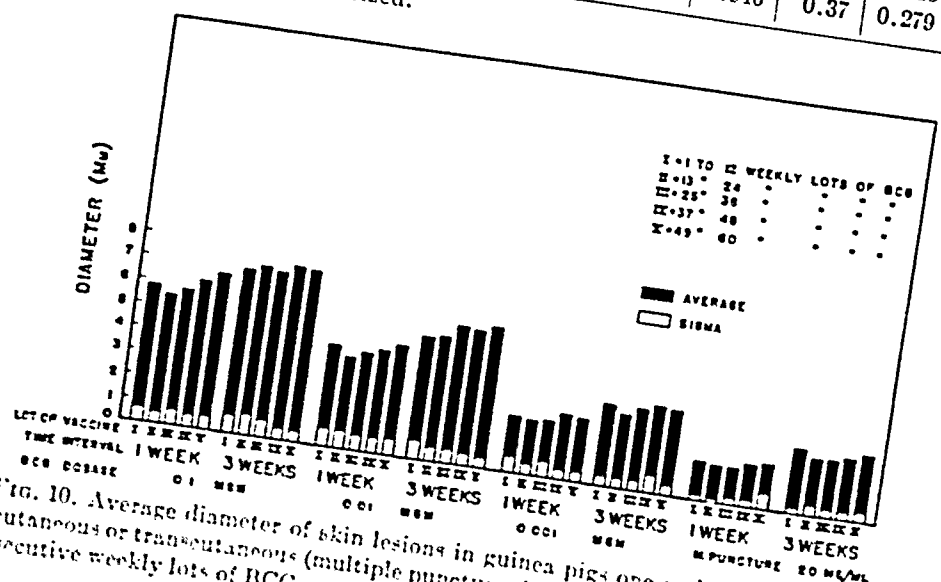

FIG. 10. Average diameter of skin lesions in guinea pigs one and three weeks after intracutaneous or transcutaneous (multiple puncture) inoculations with graded doses of sixty consecutive weekly lots of BCG vaccine.

vaccine appear to show the same trend. But only in the latter case did the deviation between the first and last weekly lots approach significance. That an assumption of increased tuberculogenic potency in the vaccine toward the end of the first year's cultivation is unfounded becomes clear by the data presented

in table 8 where the maximum skin lesions with the variously graded doses of vaccine have become remarkably stationary. The sole exception is the significant deviation in the 49 to 60 weekly lots of 0.01 mg. But this instance is not substantiated in the lesions produced by the 0.1 mg. and 0.001 mg. of the vaccine. Thus it may be concluded that the tuberculogenic potency of BCG is relatively stable for the BCG vaccine which is prepared by the standardized method. It should also be added that the tuberculin reactions, observed in these animals with graded doses of Old Tuberculin ranging from 10 to 0.001 mg., bring out the fact that the degree of tuberculin hypersensitiveness showed the same stability throughout the study. An area of induration exceeding 10 by 10 by 1.5 mm. forty-eight hours after intracutaneous injection of graded doses of tuberculin occurred as follows: one week after inoculation, approximately 150 mm.^{3} with 10 mg. OT; at two weeks, 390 mm.^{3} with 1.0 mg. OT; three weeks, 510 mm.^{3} with 0.1 mg. OT. Thereafter, until the animals were killed three and six months later, a reaction of approximately 200 mm.^{3} induration uniformly occurred with 0.01 mg. OT.

BCG'S TUBERCULOGENIC AND ALLERGENIC POTENCY IN MAN

Since January, 1947, more than 2,000 tuberculin-negative persons, with no evidence in the chest roentgenograms of disease of the lungs, have been vaccinated with the same lots of BCG vaccine. The multiple puncture method has been used except in a dozen persons injected intracutaneously with 0.1 mg. BCG. The vaccine was invariably used within one week after its preparation.

A 4 by 4 cm. piece of thin paper, sterilized for fifteen minutes at 100°C., was moistened on both sides with the 20 mg. per ml. BCG vaccine in a sterile Petri dish and placed on the ether-cleansed skin. The skin was held taut by grasping the arm or thigh on opposite sides, the head plate evenly pressed against the paper and the trigger pushed. As the needle plate descends the forty needle points become coated with vaccine and carry it into the epidermis to a depth of 1 to 3 mm. The paper is removed after one or two minutes. No bandage is necessary. Pin-point bleeding may be seen in the punctures when the skin is stretched. Excessive spontaneous bleedings should be avoided; and can be readily controlled by adjusting the extension of the needles to 2 or 3 mm. beyond the head plate. Between each vaccination, the apparatus is disinfected by immersing the head plate with the needle points protruding in boiling water from 10 to 20 seconds, followed by 10 to 15 seconds in cold sterile water. The apparatus is then placed on a pad of sterile absorbent cotton to remove excess water. A new model of the apparatus has a detachable head plate and needle plate which can be replaced with sterile parts for every vaccination. The multiple puncture vaccination causes no untoward reaction and necessitates no special care. After a few days, the skin regains its normal appearance. Suppuration of the regional lymph nodes does not occur. Within ten to twenty days the local puncture marks reappear as inflamed and infiltrated papules 2 to 3 mm. wide. These lesions cause no general symptoms. Very rarely do they become enlarged. The reactions disappear in a few weeks, and from two to three months after vaccination the process is completely resorbed without residual marks or scars. Only occasionally, faint white dots may be observed. The tuberculin test with 1.0 mg. Old Tuberculin becomes positive in nearly 100 per cent of successfully vaccinated persons 8 to 12 weeks after vaccination if visible papules have been present. A reaction is considered positive when definite induration or edema of at least 6 to 8 mm. diameter, regardless of whether redness is present or not, is present forty-eight to seventy-two hours after injection.

In the author's series of about 500 multiple puncture vaccinations between January, 1947 and May, 1948, a remarkable uniformity of the local reactions has been observed with the various lots of vaccine. Although the local reactions may be variable within one group of vaccinated persons, doubtlessly due to variations in the skin and the depth of the punctures, no really severe local reactions have been encountered. The rare instances of suppuration in individual papules have usually been associated with questionable positive-tuberculin reactors at the time of vaccination, who subsequently have presented an accelerated papular reaction (13–15).

It is too early to present data on the persistence of tuberculin hypersensitiveness in these persons. A group of 20 employees of the Division, who were vaccinated between ten and seventeen months ago, were simultaneously retested recently with 0.1 mg. OT and 0.0002 mg. PPD on the flexor surface of the left forearm. Two days later 100 per cent gave positive reactions with OT (varying between 10 by 10 and 25 by 25 mm. of induration) and only 56 per cent gave positive reactions with PPD (varying between 6 by 6 and 16 by 16 mm. induration). The same discrepancy was observed in 31 nurses who were retested with the same doses of OT and PPD one month after successful multiple puncture vaccination. After forty-eight hours 93.3 per cent gave positive reaction with OT and only 26.7 per cent with PPD, the former reactions varying between 6 by 6 and 18 by 18 mm., and the latter between 6 by 6 and 10 by 10 mm. of induration. Similar observations have been made by other physicians in New York who have also noted that, whereas the reactions with OT in BCG vaccinated persons are sharply raised, markedly edematous, and usually of two and three plus intensity, the reactions with PPD are usually flat and poorly demarcated and only slightly indurated.

Such problems as the best tuberculin test material to detect tuberculin hypersensitiveness in BCG vaccinated persons, the best method of vaccination to produce the most lasting tuberculin hypersensitiveness, and the usefulness of lyophilized BCG vaccine deserve particular consideration at present. But the subject of primary importance which this First International BCG Congress should consider is the need for a uniform technique for the production of vaccine and periodic control of its avirulence and tuberculogenic potency by exact quantitative procedures. The need for an international standard technique is very real.

SUMMARY

Uniform methods are presented for the continuous production at weekly intervals of a relatively stable and effective BCG vaccine and for periodic control of its avirulence and tuberculogenic properties.

A series of sixty consecutive weekly lots of BCG vaccine produced by such methods has been assessed quantitatively for invasiveness and the production of tissue hyperplasia in guinea pigs sacrificed three and six months, respectively, after inoculation. The data confirm that BCG's avirulence is not absolute, but is of a significantly low and stationary degree to render the animals tuberculin hypersensitive.

The same lots of vaccine have been assayed for their tuberculogenic properties, their ability to produce localized intracutaneous and transcutaneous skin lesions. The data indicate that this potency is significantly stable when the vaccine is prepared by the methods employed in this study.

Experience on more than 2,000 persons vaccinated with the material from these sixty consecutive weekly lots of BCG vaccine confirm the stability of BCG's tuberculogenic potency and relative avirulence.

An international standard technique for the preparation of BCG vaccine and for the periodic control of its avirulence and tuberculogenic potency is recommended.

SUMARIO

Estudios de la Virulencia y Tuberculogenia de Sesenta Lotes Semanales Consecutivos de Vacuna BCG Producida Conforme a una Técnica Establecida

Descríbense técnicas uniformes para la continua producción semanal de una vacuna BCG relativamente estable y eficaz y para la comprobación periódica de su avirulencia y propiedades tuberculógenas.

Una serie de 60 lotes semanales consecutivos de vacuna BCG producida con dichas técnicas fué justipreciada cuantitativamente en cuanto a facultad invasora y producción de histohiperplasia en cobayos sacrificados a los tres y seis meses, respectivamente, de la inoculación. Los datos acopiados confirman que la avirulencia de BCG no es absoluta, pero sí significativamente baja y estacionaria para convertir a los animales en hipersensibles a la tuberculina.

Los mismos lotes de vacuna fueron valorados con respecto a sus propiedades tuberculógenas, o sea su capacidad para producir lesiones intra y transcutáneas localizadas. Los datos obtenidos indican que esta potencia es significativamente estable cuando se prepara la vacuna por las técnicas empleadas en este estudio.

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A SANATORIUM PROGRAM FOR THE BACTERIOLOGICAL DIAGNOSIS OF TUBERCULOSIS[1]

The Results Obtained in Cases Admitted during a Period of Ten Years

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INTRODUCTION

In 1928, Pinner and Werner (1) pointed out that with persistent use of the available methods of sputum examination, "positive" results could be found in an extraordinarily high proportion of cases of clinically active tuberculosis. They found acid-fast bacilli in 99.2 per cent of 507 patients with active disease. They concluded that bacteriological diagnosis of active tuberculosis "...is equal to any diagnostic laboratory procedure and second to none." With the repeated use of culture and guinea pig inoculation of sputum and stomach contents, Decker, Ordway and Medlar (2) found bacilli in 67 out of 97 patients with clinically active, minimal pulmonary tuberculosis.

Diagnostic Standards, 1940 (3) includes this statement: "If sputa and gastric washings are carefully and repeatedly examined...negative results are of distinct diagnostic value. It can be safely said that a patient with demonstrable parenchymal infiltration in the lung that is apparently active, in whom tubercle bacilli cannot be demonstrated, probably has a nontuberculous pulmonary lesion." Willis and Kelly (4) investigated the value of examining the expectoration of several days pooled together. In a series of cultures, 63 per cent of all "positives" were obtained from one day specimens and 92 per cent from ten day specimens. In comparing the results from examination of two successive seven day specimens, using concentration and culture, they found that the first would fail by only 7.8 per cent to yield as many "positives" as both specimens. Their recommendation was the collection and examination of a week's total of sputum.

In 1942 the Committee on Standard Laboratory Procedure of the American Trudeau Society recommended a program for bacteriological diagnosis (5) as follows:

1. Direct smear of a morning specimen and a specimen collected over a period of twenty-four hours, or as long a time as the circumstances require. (The direct smear may be omitted if the operator chooses to do a concentration.)
2. If the direct smear is negative, concentrate the same sputum specimen.
3. Culture the same specimens after concentration if the concentration is negative, if less than 5 bacilli are found in the smear, or if there is any possible doubt whether the acid-fast bacilli are true tubercle bacilli.
4. If the first specimen is negative on microscopic examination, repeat the whole procedure on two subsequent occasions.
5. If sputum is not obtainable, gastric lavage should be performed and the sediment examined by direct smear, culture, and animal inoculation.
6. A gastric lavage is recommended in patients having little sputum or if the specimens continue to be negative.

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[1] From the Barlow Sanatorium, Los Angeles, California.

In a review of bacteriological findings on 2,248 consecutive admissions at Muirdale Sanatorium, Sanford (6) stated that in active reinfection tuberculosis direct smear and concentration tests showed an accuracy of 80 per cent. But, when cultures were also used, "only slightly less than 1 per cent of all paitents with X-ray evidence of activity could not be proved positive."

Bacteriological Program at Barlow Sanatorium

Since 1935 an attempt has been made at Barlow Sanatorium to develop a feasible program of bacteriological examinations that would yield the highest possible proportion of dependable diagnoses. While this program has changed through the years, it was developed to sufficient degree by the summer of 1936 to make it possible to present at this time the results for a period of operation of ten years, from August 31, 1936 to September 1, 1946. At the beginning of the ten year period a twenty-four hour specimen of sputum collected immediately after the patient's admission was examined by direct smear. If "negative," all of the sputum for three days was collected and examined simultaneously by concentration and culture. If the results continued "negative," the three day concentration and culture test was repeated on three separate occasions during the first month of residence and once each in the second and third months. In the second months of residence the stomach contents were aspirated and examined by culture and animal inoculation.

During the ten year period, the examination program for cases remaining "negative" became more intensive. The amount of the specimen was increased until all the sputum expectorated in six days was collected for each set of tests. The number of six day specimens was increased to the point where the sputum was collected for examination almost continuously during the first three months of residence. The number of gastric aspirations was increased to 2 and then to 3 during the first three months, one in each month. In one period all the sputum expectorated for thirty days was collected in six day batches and tested simultaneously by concentration, culture, and animal inoculation. A later development was the pooling of a six day sputum specimen with the aspirated fasting stomach contents for examination by culture and animal inoculation. The evolution of the testing program is outlined in table 1. There have been variations. This represents the average. It will be noted that from February, 1939 on, one-half to two-thirds of all the sputum expectorated during the first three months was examined.

The present program, in operation since January, 1944.

1. Immediately after admission, a twenty-four hour specimen of sputum is collected and examined by direct smear and culture[2, 3].

[2] Culture for identification purposes in case the direct smear is positive for acid-fast bacilli.

[3] A variation is the immediate collection of a forty-eight hour specimen to be examined by direct smear, concentration, and culture.

2. If the direct smear is negative for acid-fast bacilli, a six day specimen is collected and examined by concentration and culture.

3. If the concentration test shows no acid-fast bacilli, the stomach contents plus a second six day specimen are examined by culture and animal inoculation. With minimal lesions or in cases where the diagnosis of tuberculosis is particularly in question, the stomach contents are examined as soon as possible, combined with the first six day sputum specimen.

TABLE 1

Evolution of the Bacteriological Diagnostic Program 1936 to 1946

Tests carried out routinely during the first three months of residence in cases remaining "negative"

PERIOD	SPUTUM TESTS*					GASTRIC CONTENTS BY CULTURE AND GUINEA PIG
	Number			Number of days' expectoration per test	Total days' expectoration-examined	
	Concentration and culture	Concentration, culture and guinea pig	Culture and guinea pig†			
8-31-36 to 10-31-37	6			3	18	1
11- 1-37 to 1-31-39	5	1		3	18	1
2- 1-39 to 9-30-40	4	7		6	66	2
10- 1-40 to 7- 1-43	4 to 8	3		6	42 to 66	3
7- 1-43 to 1- 1-44	4 to 8		3	6	42 to 66	3
1- 1-44 to 9- 1-46	7 to 9		3	6	60 to 72	3

* The initial direct smear on a twenty-four hour specimen done in every case is not tabulated.

† These sputums were pooled and examined together with the specimens of gastric contents shown in the last column.

4. If the examinations continue to be negative, six day sputum specimens are collected continuously during the first three months of residence and examined by concentration and culture. Practically, this works out at about one specimen a week.

5. Once each month during the first three months, the aspirated stomach contents are combined with one of the six day sputums for culture and animal inoculation.

When patients continuing in residence for more than three months are still "negative," a four day sputum specimen is collected in each calendar month and examined alternately by concentration and culture and by culture only. Each third month, or oftener, gastric lavage is performed and the material, after being pooled with a four day sputum specimen, is cultured and injected into animals. In addition, search for other pathogenic microorganisms, especially fungi, is carried out.

As a general proposition, in cases once proved positive, the simplest test capable of demonstrating the bacilli is performed in each calendar month.

Notes on Technique

Collection: Sputum specimens are collected in quarter-pint, paraffin-impregnated, heavy paper containers with tight fitting covers[4]. These containers have wide mouths; they do not upset easily; they will not wet through in several days; and when the cover is in place they will not spill. They are used only once, then discarded. They are assumed to be sterile as far as tubercle bacilli are concerned. Multiple day specimens are collected two days at a time in separate containers, each container being immediately refrigerated and the portions being pooled on the morning of the test. A mimeographed sheet is sent to each new patient explaining the mechanism of raising and expectorating sputum and including directions for collection of specimens. Sometimes it is necessary for a nurse, technician or doctor to explain this personally to the patient. Each patient is required to provide a specimen of either sputum or saliva obtained by coughing or clearing the throat because some patients will deny the existence of sputum for aesthetic reasons and some are unconscious of its presence. Patients who send large amounts of saliva or sinus drainage are asked to expectorate only pulmonary sputum into their collection container while discharging excess saliva and mucous into the regular disposal cup.

Microscopic tests: Direct and concentration smears are all examined ten minutes or until found "positive." When fewer than 7 bacilli are found, a confirming test on another specimen is required for a bacteriological diagnosis.

Concentration tests are carried out by means of a flotation technique (7). Briefly, the sputum is homogenized by vigorous shaking with an equal amount of 0.5 per cent sodium hydroxide, diluted 1:10 with water and shaken again with 1 cc. of xylol. Smears are prepared from the floated surface layer that develops on standing. A picric acid counter stain is used.

In order to avoid the possibility of false positive reports due to the presence of dead bacilli from a previous test, the glassware is baked at 250° C., a temperature high enough to carbonize organic matter, or treated with sufficiently strong cleaning acid to destroy the bacillary bodies.

Concentration tests are always accompanied by culture, carried out simultaneously on the same specimen after homogenization.

Culture: The sputum-sodium hydroxide digest, prepared as above, is mixed with an equal amount of 2.5 per cent oxalic acid, centrifuged, and the sediment planted on 2 tubes of Petragnani's medium[5], 2 drops to a tube (8). The culture tubes are discarded at two months.

Animal inoculation: The sputum-sodium hydroxide digest, as above, is washed once and the sediment inoculated subcutaneously into the lower flank of a guinea pig. Animals with clinical signs of disease are autopsied at one month, others at two months. Visceral disease and the demonstration of acid-fast bacilli are required for the report of a positive test (10).

Gastric tests: Fasting stomach contents and washings with or without added sputum are made up to a final concentration of 0.5 per cent sodium hydroxide (by addition of 4 per cent sodium hydroxide in the amount of 1/8 the volume of the specimen), shaken, and the sediment divided for culture and animal inoculation as above (10). Four tubes of culture medium are planted with sediment from the sodium hydroxide digest, an equal number with that from the oxalic acid-treated material. (In cases where the culture is positive and the guinea pig negative, the cultural growth is inoculated to prove its virulence.)

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[4] "White-tite" containers manufactured by Fibre Board Products, Inc., Stockton, California; or "Seal-Right" containers manufactured by Seal Rite, Inc., Fulton, New York.

[5] Actually at present, 2 tubes of Lowenstein's (Jensen modification), 2 of yolk-potato medium, as recommended by the Committee on Laboratory Procedure of the American Trudeau Society (9), and 2 of Petragnani's medium.

Microscopic examinations are never done on stomach contents since, although the stomach tubes and other equipment are sterilized, dead tubercle bacilli from a previously positive case may adhere to the tube or syringe and give a microscopically false positive result.

Clinical Evaluations

The clinical evaluation of each case was considered by the clinical staff headed by Dr. Howard W. Bosworth, including interpretations of the roentgenograms by Dr. Rolla G. Karshner, and final designations by Dr. Franklin S. Reding. The cases were classified[6] as to the presence of activity and according to the stage of the disease. Active disease was diagnosed when the admission roentgenogram showed characteristic soft shadows and when successive pictures over a period of months or years showed unquestioned changes. The presence of signs and symptoms not explained by other conditions was given due weight. Finally, and possibly at variance with other clinicians, all cases bacteriologically positive were considered "active." This was so even in those positive only by culture and animal inoculation.

TABLE 2

Clinical Classification

Patients admitted from August 31, 1936 to September 1, 1946

CLASSIFICATION	ALL ADMISSIONS		PATIENTS RESIDENT TWO MONTHS OR MORE	
	Number	Per cent of total	Number	Per cent of total
Minimal	172	24.2	166	25.6
Moderately advanced	297	41.7	277	42.9
Far advanced	129	18.1	126	19.4
Pleural tuberculosis*	12	1.7	10†	1.5
No active tuberculosis	45	6.3	38	5.8
No tuberculosis	56	7.9	31	4.8
No diagnosis, no treatment	1	0.1		
Total	712	100.0	648	100.0

* No pulmonary tuberculosis demonstrated.

† One case subsequently shown to be neoplastic in origin.

RESULTS

During the ten year period from August 31, 1936 to September 1, 1946, 712 patients were admitted to Barlow Sanatorium. Of these, 648 remained two months or more. The numbers and percentages of the different clinical categories are shown in table 2.

Table 3 shows the numbers and percentages of cases found bacteriologically positive, according to the clinical designation, for those resident two months or more. Of the 569 cases classified as active pulmonary tuberculosis, pulmonary

[6] According to Diagnostic Standards, 1940, National Tuberculosis Association.

secretions were "positive" in 97.4 per cent. Including pleural fluid examinations, bacteriological diagnoses were obtained in 97.7 per cent. Positive results were obtained in 94.6 per cent of cases of minimal tuberculosis.

Of the "positive" cases recorded in table 3, there were 13 positive by one test only: direct smear (Gaffky III) in one case, culture in 4 and animal inoculation in 8. There were 18 cases positive by microscopic tests only: all were Gaffky II or better; in 17 there were 2 or more positive tests; in 16 there were 2 or more tests of Gaffky III or better. In no case was a positive diagnosis dependent on a plus one[7] or a Gaffky I microscopic reading. In only one case was a bacteriological diagnosis made from the concentration test alone. Here there were 6 positive tests varying from plus one to plus four.[8]

TABLE 3

Bacteriological Diagnoses

Patients admitted from 8–31–36 to 9–1–46 who were resident two months or more

CLINICAL CLASSIFICATION	NUMBER OF PATIENTS	BACTERIOLOGICALLY POSITIVE	
		Number	Per cent
Minimal	166	157	94.6
Moderately advanced	277	272*	98.2*
Far advanced	126	125†	99.2†
Pleural effusion, no demon. pulmonary tuberculosis	10	2	20.0
No active tuberculosis	38	0	0
No tuberculosis	31	0	0
Total	648	556	85.8
Total active pulmonary tuberculosis	569	554‡	97.4‡

* 273 and 98.5 per cent including 1 case with "negative" sputum but "positive" pleural fluid.

† 126 and 100 per cent including 1 case in which the pleural fluid was "positive."

‡ 556 and 97.7 per cent including 2 cases with "positive" pleural fluid.

Patients bacteriologically negative with clinically active pulmonary tuberculosis: Among those resident two months or more there were 15 such cases. Eight had minimal; 6, moderately advanced and 1, far advanced tuberculosis. Pertinent data for each case are shown in table 4.

It will be noted that in 2 cases artificial pneumothorax had been instituted before admission. Although the pulmonary disease was undoubtedly active in one and probably active in the other, access to drainage was apparently blocked. But in both cases tubercle bacilli were found in the pleural fluid and, in a sense, a positive bacteriological diagnosis could be made.

In a third case lobectomy had been performed prior to admission. Here a clinical diagnosis of minimal tuberculosis was made on the theory that non-demonstrable active disease must exist in the other lobes.

Of the remaining 12 "negative" cases, clinical review indicated 3 to be un-

[7] One to 6 acid-fast bacilli in ten minutes search.

[8] More than 2 bacilli per field.

questionably "active," *all* with *minimal* lesions. One of these had had rest in bed for eight weeks, and another for twelve weeks, before admission. In the experience of the staff this sometimes is sufficient to result in "closure" of the lesion though not sufficient to allow for complete healing. Among 6 cases rated as probably "active," 4 had had rest in bed for eight weeks or more prior to admission.

TABLE 4

Cases Clinically Active and Bacteriologically Negative

Resident two months or more

CASE	CLINICAL CLASSIFICATION	MANTOUX	PLEURAL FLUID	TREATMENT BEFORE ADMISSION		CLINICAL ACTIVITY REASSESSMENT
				Weeks in bed	Surgical	
1	MA	Positive		8		Probable
2	Min.	Positive				Probable
3	Min.	Positive		10		Probable
4	Min.	Not done	Yes	1	Lobectomy	Questionable
5	Min.	Positive		8		Unquestionable
6	Min.	Positive				Probable
7	MA	Positive		1		*Question as to* tuberculosis
8	MA	Positive		8		Probable
9	Min.	Positive		3		Unquestionable
10	Min.	Positive		12		Unquestionable
11	Min.	Positive		2		Probable
12	MA	Positive	Tubercle bacilli	100	Pneumothorax	Probable
13	MA	Positive		0		Questionable
14	MA	Positive				Questionable
15	FA	Positive	Tubercle bacilli	8	Pneumothorax	Unquestionable

Review showed one case in which the diagnosis of tuberculosis could be questioned. The disease was limited to a thin-walled cavity that disappeared on bed rest. Examination for fungi had not been made.

Comparison of results in the early and late part of the study: The results of the bacteriological study are derived from a program that has become increasingly elaborate during the ten year period. The minimum schedule of examinations that will yield comparable results is not indicated. Table 5 shows the results for the first two years and the last two years of the study. There is very little difference in the percentage of positive findings (for cases of active pulmonary tuberculosis) in spite of the more extensive examination program carried out in the latter period. This is perhaps explainable by the fact that in the two years, 1936 to 1938, 8.2 per cent of the admissions (remaining two months or more) were mini-

mal cases and 34.4 per cent, far advanced; and in the two years, 1944 to 1946, 34.5 per cent of the admissions were minimal and only 14.7 per cent were far advanced cases.

DISCUSSION

The program for bacteriological diagnosis presented may be thought too elaborate for many institutions. It is not known how much it could be reduced without materially affecting the results. But as it stands it is well worth the cost in the resultant high caliber of clinical diagnosis and treatment.

There is much room for improvement in the program. Better culture media and culture methods are developing. More frequent gastric lavage is indicated. But the success of any program depends on the development of as fool-proof a routine as possible, constant attention, vigilance and frequent reassessment of methods.

In the present classification all bacteriologically positive cases have been considered as having active disease regardless of other clinical findings. How does

TABLE 5

Comparison of Results in the Early and Late Part of the Study

Active cases of pulmonary tuberculosis resident two months or more

TWO-YEAR PERIODS	ADMITTED	BACTERIOLOGICALLY POSITIVE	
		Number	Per cent
1936 to 1938	99	96	96.9
1944 to 1946	123	120	97.6

this affect the results? If the clinically nonactive, bacteriologically positive cases were eliminated from the totals, the percentage of "negative" results would be somewhat raised.

It may be questioned whether it is possible to have any cases with clinically active disease that are completely bacteriologically negative. With an improved examination program, surely the proportion of "negative" cases will be reduced. In cases of strictly productive disease with well walled off tubercles or of well encapsulated caseous lesions, and in those in which the draining bronchus has been closed by collapse therapy or otherwise, no output of tubercle bacilli would be expected. The "closed" but "active" case is theoretically possible and does exist in practice.

It is believed that all cases rated bacteriologically positive are dependably so. A false positive result is considered to be far worse than failure to find tubercle bacilli that *are* present. As mentioned, the finding of rare bacilli on the microscopic preparations has not been accepted as a bacteriologically positive result. Concentration results have been checked by culture and questionable cultural growths by animal inoculation. The reading of microscopic, cultural, and animal tests has been closely supervised.

In comparing the results of different bacteriological programs it is important to know the clinical classification of the group studied. The percentage of positive bacteriological findings in a given series of patients will vary according to the proportion with advanced and minimal lesions. In the present series the proportion of minimal cases is unusually high, making up approximately one-quarter of the total, and that of the far advanced cases rather low, being only one-fifth of the total.

SUMMARY

1. An intensive program for the bacteriological diagnosis of tuberculosis is presented.

2. Where indicated, the program includes examination of two-thirds to four-fifths of all sputum expectorated by a patient during the first three months of residence by concentration and culture and, partly, by animal inoculation. In addition, stomach contents are examined by culture and inoculation once each month.

3. The evolution and use of this program during a ten year period is outlined.

4. During the ten year period positive bacteriological results were obtained in 97.4 per cent of clinically active cases of pulmonary tuberculosis resident two months or more.

5. Positive results were obtained in 94.6 per cent of cases of clinically active *minimal* tuberculosis resident two months or more.

SUMARIO

El Diagnóstico Bacteriológico de la Tuberculosis en un Sanatorio

1. El plan expuesto contempla el diagnóstico bacteriológico de la tuberculosis en forma intensa.

2. Siempre que esté indicado, el plan comprende exámenes de dos tercios a cuatro quintos de todo el esputo expectorado por un enfermo durante los primeros tres meses de su estancia en el sanatorio. Los exámenes comprenden concentración y cultivo del esputo y, en algunos casos, inoculación en animales. Además, se examina una vez al mes el contenido gástrico por medio de cultivos e inoculación.

3. Bosquéjanse la evolución y el empleo de este sistema durante un decenio.

4. Durante el decenio del estudio obtuviéronse resultados bacteriológicos positivos en 97.4 por ciento de los casos clínicamente activos de tuberculosis pulmonar en residencia dos meses o más.

5. Obtúvose igualmente resultado positivo en 94.6 por ciento de los casos de tuberculosis *minima* clínicamente activa que habían residido en el establecimiento dos meses o más.

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LETTER TO THE EDITORS

USE OF A SLIDE CULTURE TECHNIQUE FOR RAPID STREPTOMYCIN SENSITIVITY TESTING

To the Editors of the American Review of Tuberculosis:

We have found it possible to perform streptomycin sensitivity tests employing a modification of the slide-culture method first described by Pryce. (Pryce, D. M., J. Path. Bact., 1941, *53*, 327.)

For the past year we have been comparing this method with standard techniques of performing sensitivity tests with most promising results.

The method is rapid, easily performed, and is carried out as follows:

Pure cultures, or digested neutralized specimens, are smeared thinly over the surfaces of 5 sterile 3 by ½ inch glass slides (made by cutting a 3 by 1 microslide longitudinally). Employing sterile slide forceps, the smear preparations are immersed into a series of tubes of Tween-albumin medium. (Dubos, R. J., Proc. Soc. Exper. Biol. & Med., 1945, *58*, 361) containing 0, 0, 2, 10, and 100 γ of streptomycin.) Other suitable liquid media may be employed.

After incubation at 37°C. for one week, a smear is removed from a control tube (no streptomycin) and is stained by the usual Ziehl-Neelsen technique. The preparation is examined by low power microscopy and, if colonies with serpentine cords (Middlebrook, G., Dubos, R. J., and Pierce, C., J. Exper. Med., 1947, *86*, 175) are seen, the remainder of the smears are also removed, stained, and examined in a similar manner. The concentration of streptomycin which inhibits growth on the slide is considered the end point.

The sensitivity readings compare favorably with tests made, using more standard methods employing inspissated egg media and egg-yolk-agar medium. It was interesting to note that one could estimate "partially resistant" strains by comparing colony size and numbers of colonies with the growth on the control tube.

We have also attempted to utilize this technique by smearing selected particles from pathological material directly on the slide and decontaminating the preparation by immersing it in 5 per cent H_2SO_4 or 3 per cent NaOH. Although positive results have been obtained, they have been too erratic to recommend this procedure unless duplicate series of smears are made.

It is our hope that other laboratories will evaluate this technique as a rapid method for testing the sensitivity of *M. tuberculosis* to streptomycin or other antibacterial agents.

U. S. Public Health Service
Tuberculosis Evaluation Laboratory
Atlanta, Georgia.
March 15, 1949

Martin Cummings,
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THE

AMERICAN REVIEW OF TUBERCULOSIS ABSTRACTS

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Childhood Tuberculosis.—Most reports of tuberculosis in children confine themselves to a study of the primary complex. The disease may travel diverse paths, however, and the character may be protean. A study of 90 cases which came to autopsy at Sea View Hospital is reported. All the patients were less than 13 years of age, and 40 per cent less than 3. There were 67.8 per cent Negro children in the series, a reversal of the over-all ratio in the hospital. The reason would seem to be the presence of a low resistance and consequent overwhelming infection in this group, with frequent progressive primary complexes and dissemination. There were 3 general types of disease: the primary complex, hematogenous dissemination, and chronic pulmonary tuberculosis. The frequency of various lesions was as follows: (*1*) Healed or healing primary complex occurred in 66 cases. (*2*) Progressive primary complex was seen in 23 cases. The lymph nodes were often grossly involved, both locally and in distant sites. (*3*) Early generalized hematogenous tuberculosis was present to some extent in all 23 cases of progressive primary complex. (*4*) Late generalized hematogenous lesions occurred in 6 cases, all of which had active extrapulmonary disease. (*5*) Chronic pulmonary tuberculosis was present in 20 cases. This incidence in children (22 per cent of the series) is notably higher than in other reports. The lesions were similar clinically and roentgenologically to lesions in older age groups. A difference at autopsy was the frequency (7 cases) of caseated regional nodes in children, a rare finding in chronic pulmonary lesions of adults. (*6*) Skeletal tuberculosis was present as an active disease in 21 cases. All of the 13 cases with vertebral lesions had associated abscesses. All but one of the cases of bone disease came from progressive primary lesions. (*7*) Genito-urinary lesions were present in 10 cases, 9 of which were females. Three of the cases had renal lesions; 8 had genital lesions, chiefly of the fallopian tubes. (*8*) Thirty-seven cases of fatal tuberculous meningitis were included, a far greater incidence than in adults. Tuberculomas of the brain were present in 78 per cent of these. The sources of the brain lesions were fresh primaries in 5 cases, progressive primaries in 12, healed primaries in 17, and active extrapulmonary disease in only 3. (*9*) Two cases of extensive intestinal ulceration without pulmonary cavitation were found. (*10*) One case of tuberculous valvular endocarditis was noted. This is so rare that it may be the eighth case described in the literature.—*Tuberculosis in children, O. Auerbach, Am. J. Dis. Child., April, 1948, 75: 555.*—(*W. H. Oatway, Jr.*)

Phthisiogenous Infiltrates.—Laennec, Koch and their contemporaries believed that pulmonary tuberculosis started in the apices and spread apicocaudally. This conception was maintained into the Nineteen-twenties. In 1922 Assman demonstrated a form of isolated tuberculosis in adults which Simon named *"Frühinfiltrat"*. Braenning and Redeker developed the thesis that this is a clinical-roentgenological entity and concluded that most tuberculosis originated from these infraclavicular "early infiltrates" which spread

not apicocaudally but in periodical propagations. During the past twenty years this doctrine of "early infiltrates" has been shown to be essentially wrong. Scandinavian researches notably by Heimbeck, Scheel, Malmros and Hedvall and Frostad showed that some of these "early infiltrates" were primary infiltrates. They also found that pulmonary tuberculosis originated from "subprimary" initial foci situated in the supraclavicular region or first intercostal space or in both regions twelve to fourteen months after the primary infection. The variation is one of time, acute as contrasted with subchronic or chronic development after a primary infection. Following BCG vaccination, a typical "early infiltrate" may appear in the lung. Pulmonary tuberculosis may develop from hematogenous foci after secondary spread, from isolated hematogenous infiltrates, and finally from exogenous or endogenous reactivated infiltrate-residues. Thus destructive phthisis may develop from infiltrates or residues of infiltrates which differ as to genesis, aspect and age. These are all phthisiogenous forms of infiltrates.—*On the different forms of phthisiogenous infiltrates and the development of phthisis, N. Christiansen, Acta radiol., 1948, 30: 17.*—(*J. E. Farber*)

Renal Tuberculosis.—A tuberculous stricture of the ureter is most likely to occur at (*1*) the uretero-pelvic junction, (*2*) the juxtavesical portion or (*3*) the point where it crosses the pelvic brim. The formation of a stricture may lead to abatement of vesical symptoms as the result of one of the following changes: (*1*) tuberculous hydronephrosis, (*2*) extensive renal caseation with or without infiltration of calcium salts, or (*3*) fibrosis, the so-called "autonephrectomy". The first two types are sources of danger by rupture, secondary infection, or miliary tuberculosis. The three types are seen in 20 per cent of all cases of renal tuberculosis; type 3 alone is seen in 0.5 per cent. Calcification is not *per se* a favorable sign. With an obstructed ureter, kidney disease may not be suspected so that other abdominal conditions may be considered first or evidence of the condition may be noted in a routine investigation of a case of tuberculosis. In the roentgenogram, the kidney outline is enlarged, with or without intrarenal or ureteral calcification. The intravenous pyelogram shows nonfunction of the kidney. On cystoscopic examination, the ureteric orifice is puckered and scarred; it may be difficult to pass a catheter. The treatment is surgical removal of the kidney and ureter. If the condition appears quiescent, operation may be deferred; clinical criteria are more important than roentgenographic findings in this decision.—*Silent renal tuberculosis, J. H. Carver, Tubercle, December, 1948, 29: 269.*—(*A. G. Cohen*)

Extrafascial Pneumothorax.—In certain cases, after a first stage thoracoplasty with Semb apicolysis, complications arise which delay or prevent the performance of additional stages. In such cases it is desirable to retain the benefits of the first stage. In other cases, with small apical cavities, a three-rib thoracoplasty suffices. In both situations, the author used air to keep open the Semb space. Insufflations were begun during the second postoperative week. Refills were continued for six months, by which time the regenerated ribs could keep the apex down. In some cases, oil was substituted for air. In a series of 31 cases, 7 were unsuccessful due to various complications. In the others, the original collapse was maintained until and after the space was allowed to obliterate. Thus additional stages were delayed safely. In 9 patients with small apical cavities, additional stages were not required.—*Extrafascial pneumothorax, M. Konstam, Thorax, December, 1948, 3: 247.*—(*A. G. Cohen*)

Hemorrhage in Extrapleural Pneumothorax.—Calcium alginate was found experimentally to possess hemostatic properties. In the operation for extrapleural pneumothorax, the walls of the space were painted with a 4 per cent solution of sodium alginate; this was followed by application of 2 per cent calcium chloride. The procedure appeared to control

hemorrhage effectively without untoward effects.—*The control of haemorrhage in extrapleural pneumothorax, K. S. Mullard, Thorax, December, 1948, 3: 288.*—(*A. G. Cohen*)

Artificial Pneumoperitoneum.—Peritoneal effusion is the most common of the infrequent complications of artificial pneumoperitoneum used in the treatment of pulmonary tuberculosis. It has occurred in 8 (3.58 per cent) of 223 cases who had received 5,800 refills. The ascites in these cases is often due to tuberculous peritonitis. Its incidence is unrelated to the activity of the pulmonary disease. Adhesions appear early but the response to laparotomy is fairly good. The appearance of ascites is considered an indication for discontinuation of the pneumoperitoneum.—*Peritoneal effusion as a complication of artificial pneumoperitoneum, R. C. Cohen, Lancet, December 25, 1948, 2: 1006.*—(*A. G. Cohen*)

BCG Vaccination.—Ninety per cent of infants from homes with infectious tuberculosis in Dublin become infected during the first year of life. Autopsy evidence shows that most deaths resulted from direct spread from the unhealed primary complex. In more recent years, as the result of better and more prompt treatment, there were more recoveries; still, in the best year at least 41 per cent died. Sixty infants under the age of one year were vaccinated with BCG and then returned to tuberculous homes. They were followed for one to ten years. There were no deaths from tuberculosis.—*The need for BCG vaccination in infants, D. S. Price, Tubercle, January, 1949, 20: 11.*—(*A. G. Cohen*)

BCG Vaccination.—There is a very high incidence of tuberculosis among the Saskatchewan Indians. For a period of about thirteen years, alternate infants were given BCG vaccination. A total of 306 infants were vaccinated and 303 were followed as controls. The alternation was arranged in such a fashion as to insure comparability of the vaccinated and control groups. Vaccinations were performed within ten days of birth. In all but 21 cases, the intracutaneous route was used. Approximately one-half of the infants in each group were in close contact with infectious adult tuberculous patients. Follow-up data were available for almost all cases. In the vaccinated group, there were 6 cases of tuberculosis (1.96 per cent) with 2 deaths. In the control group, there were 29 cases of the disease (9.5 per cent) with 9 deaths. The ratio of incidence of infection in the 2 groups was 1 :4.85; the ratio of deaths was 1 :4.5. There were no untoward effects from the vaccination. —*BCG vaccination of Indian infants in Saskatchewan, R. G. Ferguson & A. B. Simes, Tubercle, January, 1949, 30: 5.*—(*A. G. Cohen*)

BCG Vaccination.—In autopsies performed upon children who died of contagious diseases, special studies were made of the various chains of lymph nodes and also of the tonsils, liver, kidneys, spleen and lungs. The subjects included 57 children under the age of two years who had been vaccinated with BCG-Paris and 25 controls. Characteristic changes were found up to three months after vaccination. The lymph nodes contained large dilated sinuses filled with large endothelial cells and lymphoblasts. Reticulum cells were scarce; monocytes and histocytes were absent. The afferent lymphatics were filled with a pale, rose-colored homogeneous substance. The lungs showed accumulations of histiocytes and lymphocytes. The interalveolar and interlobar septa and pleura were thickened and edematous and the alveoli were distended. The spleen contained hyperplastic follicles and endothelial proliferation. Round cell infiltrations of the liver were noted. In the group vaccinated three to six months previously, the changes were less marked. After one year, there was progressive fibrosis of the lymphatic apparatus. After two years, the nodes contained increasing young connective tissue elements while fibrous tissue was more pronounced at

the periphery. Fibrous strands projected radially into the gland substance. The lungs, liver and spleen showed few changes. It was concluded that the cellular reaction to BCG starts in two to three weeks, reaches its height in the third month and then declines. At the end of one year there is beginning progressive fibrosis.—*Pathologic changes in children vaccinated with BCG, V. I. Puzik, Amer. Rev. Soviet Med., October, 1948, 5: 189.*—(*A. G. Cohen*)

Vitamin A for Skin Tuberculosis.—In 45 patients with skin tuberculosis a low vitamin A level in the blood was found in association with a high carotene content. Administration of 50,000 units of vitamin A daily for three to four months produced normal values in the blood. Epithelization of ulcerations and absorption of lupus infiltrates was observed in 27 patients treated with this dosage. Improvement was noted in 21 of the 27 patients, 16 showing marked improvement and 5 moderate improvement. No clinical cure was achieved in lupus whereas 4 of 6 cases of scrofuloderma appeared clinically healed. The therapeutic results of vitamin A were enhanced if combined with vitamin C administration.—*Vitamin A and its importance in skin tuberculosis (Russian), V. L. Althausen and M. V. Lepskaia, Probl. Tuberk., 1948, 1: 57.*—(*V. Leites*)

Calciferol for Lupus.—Fifty cases of lupus were treated with calciferol. In 38 of these cases the disease had lasted from five to fifteen years, in 12 from twenty to thirty-five years. The daily dosage was 100,000 to 200,000 international units in an alcoholic solution and was given for four to six months. Of the 50 patients, 33 appeared clinically cured, 6 were markedly improved, and 11 moderately improved. The patients in the latter two groups were still being treated. The stability of the results and the incidence of relapse must be evaluated later but biopsy specimens of the skin and mucosa in cases appearing clinically cured showed anatomical healing in 3 of the 11 cases examined. All other speciemens revealed persistence of tubercles and giant cells. Complicating gastro-intestinal and urinary disturbances were found in 40 per cent of the cases but they were of a mild and transitory nature.—*Treatment of lupus with vitamin D_2 (Calciferol) (Russian), A. B. Weinstein, V. L. Althausen, & A. R. Tatianin, Probl. Tuberk., 1948, 5: 31.*—(*V. Leites*)

Vitamin D in Sarcoidosis.—Because favorable results have been obtained with vitamin D in the treatment of lupus vulgaris, the lesions of which resemble sarcoidosis, vitamin D therapy was tried in the latter. A case is reported of a woman who was known to have had sarcoidosis of the skin of the face for nine years with no remissions and whose chest roentgenograms showed prominent hilar shadows. She did not react to the first Mantoux test; on retesting there was a reaction at 72 hours although there was none at 48 hours. She was given 100,000 units of calciferol daily by mouth for fifteen days, when toxic symptoms developed and the treatment was stopped. There was no rise in the serum calcium at this time but the blood urea nitrogen rose to 60 mg. per 100 ml. There was dramatic improvement of the skin lesions. She later had another course of calciferol 50,000 units every second day, and again became intolerant. The clinical improvement remained and it is believed that the drug favorably influenced the sarcoidosis of the face. There was slight progression of the cystic changes of the fingers, however, and the roentgenographic appearance of the chest was not altered; her bronchitic symptoms became worse. Calciferol should not be used in the treatment of sarcoidosis unless the skin lesions are of sufficient severity to justify the risk of these untoward reactions.—*Vitamin D in treatment of Boeck's sarcoidosis, R. F. Robertson, Brit. M. J. December 18, 1948, 4589: 1059.*—(*R. W. Clarke*)

Roentgenotherapy of Sarcoidosis.—Sarcoidosis runs a chronic, relapsing course often with mild symptoms but sometimes

with great damage to many structures. Many forms of treatment, including roentgenotherapy, have been unsuccessful. The authors report 11 cases in which they employed roentgenotherapy to various areas, including the skin, eyes, lymph nodes, and lungs. In one patient, skin nodules disappeared after doses of 3,000 and 2,500 r. but another nodule in the same person did not regress with 2,000 r. The lesions of one patient regressed spontaneously eleven months after irradiation was completed.—*X-ray therapy of Boeck's sarcoid, C. P. Donlan, Radiology, August, 1948, 51: 237.—(G. F. Mitchell)*

Heterospecific Therapy for Tuberculosis.—Experiments begun in 1924 demonstrated the production of a "heterospecific increase of resistance" when animals were inoculated with vaccines prepared from a variety of common bacteria. Rabbits and guinea pigs were more resistant than usual to pathogenic bacteria, including *Mycobacterium tuberculosis* for five to twenty days after intravenous or subcutaneous inoculation with dead bacteria. The greatest protection was obtained by using a mixture of heat-killed *E. typhosa* and *N. gonorrhoeae* in a ratio of 1:3. *E. typhosa* is said to stimulate the function of the reticulo-endothelial system and *N. gonorrhoeae* to produce an infiltration of plasma cells. Weekly subcutaneous inoculations with this mixture partially protected the animals against subsequent injections of tubercle bacilli and favorably altered the course of an established tuberculous infection. A total of 283 tuberculous patients were treated with the vaccine prior to 1937 and the results were considered "favorable." In 1940 a mixed autolysate of *E. typhosa* and *N. gonorrhoeae* ("Heterosate") was found to be more effective than the vaccines in the prevention and treatment of experimental tuberculosis in animals. From 1940 to 1945, 932 tuberculous patients were treated with "Heterosate." In preparing this material a clear autolysate was obtained from a suspension of 2.0 mg. of bacteria per cc. The autolysates from *E. typhosa* and *N. gonorrhoeae* were then mixed in a 1:3 ratio. Weekly subcutaneous inoculations were begun with 0.25 cc. of a 1:100,000 dilution and the inoculum was increased over a period of thirty weeks to 1.0 cc. of undiluted "Heterosate." The course of treatment consisted of 36 to 176 inoculations. Of 300 patients with "incipient" tuberculosis, 65 improved and 235 completely recovered; of 403 "light cases", 86 improved and 315 recovered. The course of the disease was not changed in 2 "light cases" with pleurisy. "Moderately severe" disease was improved in 88 and cured in 65 of 173 patients; 18 were unchanged and 2 died. Of 56 "serious cases", 19 improved, 7 recovered, 25 were unchanged, and 5 grew worse. Prophylactic inoculations of "Heterosate" apparently afforded some protection against tuberculosis in a series of 44 student nurses.—*Heterospecific alteration therapy for pulmonary tuberculosis, S. Nukada & C. Ryu, Japanese Med. J., June, 1948, 1: 181.—(L. B. Hobson)*

Congenital Pneumonia.—An epidemiologic and serologic relationship exists between the occurrence of simple upper respiratory infection and primary atypical pneumonia. Primary virus pneumonitis occurs in epidemics attacking newborn and premature infants. Surface cells from the pharynx and lungs often contain cytoplasmic inclusion bodies but bacteria are scanty. Treatment is ineffective. Six instances are reported in which infants were born with respiratory symptoms or developed them during the first or second day. The mothers had symptoms of respiratory infection before or after childbirth. Both infants and mothers were found to have inclusion bodies in the pharyngeal smears. Normal human serum, U.S.P., and concentrated human gamma globulin appeared to be beneficial for the infants' infections.—*Congenital pneumonitis in newborn infants, J. M. Adams, Am. J. Dis. Child., April, 1948, 75: 544.—(W. H. Oatway, Jr.)*

Lungs in Scleroderma.—In 4 cases of

generalized scleroderma, chest roentgenograms showed diffuse, irregular, fibrotic lesions throughout both lower lung fields. In these, as well as in most previously reported cases, there were no pulmonary symptoms aside from progressive exertional dyspnea. The changes are probably due to increase in pulmonary connective tissue. The pulmonary condition may precede the appearance of skin changes. It may be found also in patients with the Raynaud phenomenon but without scleroderma.—*Pulmonary fibrosis in generalized scleroderma, W. E. Lloyd & R. D. Tonkin, Thorax, December, 1948, 3: 241.—(A. G. Cohen)*

Atypical Pneumonia.—Between March, 1945 and March, 1946, the authors saw 49 cases of lobar pneumonia of which less than 10 per cent showed the typical picture. In the other cases the disease began gradually with a slow rise of temperature and subfebrile temperatures were observed through many weeks. Repeated episodes of slight fever, however, were common. Bronchial breathing was hardly ever heard but crepitant rales were present for a long time. Herpes labialis was rare. The leucocyte count was rarely more than 10,000. In 26.4 per cent of the patients, roentgenologic examinations showed pulmonary infiltrations present after four weeks. There was no difference in the course of the disease in 25 patients treated with sulfonamids as compared to that in 24 patients who received the old treatment with quinine and camphor. There was a mortality of 32 per cent. It is believed that the atypical course in these patients was due to their poor nutrition, especially the lack of protein and vitamins in their food.—*Uber die besonders Verlaufsform der Pneumonien des letzten Jahres, W. Groeger & E. Treml, Wien. klin. Wchnschr., December 5, 1947, 59: 794.—(G. C. Leiner)*

Pulmonary Hemosiderosis.—The rare disease of pulmonary hemosiderosis was first described in 1921. The patients have cyanosis and dyspnea in early life with a cough productive of a blood-tinged sputum containing hemosiderin. Anemia, eosinophilia and leukopenia may appear. Fever and an elevated sedimentation rate also occur. Physical examination reveals dulness over the lung, harsh breath sounds, and rales. The roentgenographic picture is pathognomonic, with diffuse shadows scattered more or less over both lung fields, absolutely independent of the boarders of the lobes and sometimes denser toward the bases. This cloudiness, however, often alternates with a mossy mottling. In the early stages mottling is a dominating feature. The hilar shadows are enlarged and deepened. The alveoli are more or less filled with iron pigment, thus reducing the air content. Treatment is symptomatic and death is the final outcome in spite of remissions.—*Hemosiderosis of the lungs—Typical roentgenological findings, O. Elgemark & S. R. Kjellberg, Acta radiol., 1948, 29: 82.—(J. E. Farber)*

Pulmonary Manifestations of Leukemia.—A case of myelogenous leukemia in a 3 year old girl is reported. The respiratory symptoms were predominant in the clinical picture and consisted of dyspnea, cough, bloody expectoration and fever. Roentgenograms showed diffuse mottling in both lung fields. The tuberculin test was negative. At autopsy the lungs were generally congested with numerous infarcts and intra-alveolar edema. The interalveolar septa were extensively infiltrated with leucoblastic and monocytic elements.—*Leucémie aiguë à forme pulmonaire chez une fillette de 8 ans, R. Clément et R. Gormezano, Presse méd., June 18, 1948: 446.—(V. Leites)*

Retrosternal Infiltration in Malignant Lymphoma.—There are no dependable pathognomonic roentgenographic signs of intrathoracic lymphomas and they may assume many different forms. The authors find that, in addition to the six types generally described, there is also a retrosternal type occurring alone or with one of the other forms. This finding may be correlated with the anatomic distri-

bution of lymphoid tissue in the retrosternal area. Enlargment of the hilar or mediastinal lymph nodes usually is looked for when a clinical diagnosis of malignant lymphoma is made, but the other manifestations may be neglected. The authors have found a lateral roentgenogram of the chest helpful in demonstrating (*1*) a "boardlike" soft tissue mass of even width extending from level of diaphragm to that of the sternoclavicular joint, or (*2*) a "padlike" mass or masses with a lobulated appearance and a broad base toward the sternum.—*Retrosternal infiltration in malignant lymphoma, F. G. Fleischner, C. Bernstein & B. E. Le Vine, Radiology, September, 1948, 51: 350.—(G. F. Mitchell)*

Peripheral Vascular Diseases of the Lung.—Various disease states affect the smaller pulmonary blood vessels by increasing the permeability of their walls or by causing obstruction of their lumina. The authors are concerned with the roentgenographic changes which are seen in diseases affecting vascular permeability. This permeability may be disturbed by (*1*) trauma which may alter the vessel wall, rupture it, or produce prolonged arteriolar spasm, (*2*) chronic nutritional deficiency, or (*3*) exposure to drugs or antigenic substances. Nonpenetrating injuries of the chest frequently produce roentgenographic signs suggestive of an acute inflammatory process. These include parenchymal shadows of variable extent usually associated with pleural involvement and tending to disappear rapidly. Epidemic influenza damages the pulmonary capillaries primarily, producing a progressive hemorrhagic pneumonia with shadows rapidly extending from the periphery toward the hilum. Disseminated lupus erythematosus shows a roentgenographic picture similar to that in influenza but is, of course, characterized by diffuse fibrinoid degeneration of collagen with necrotizing arteritis. Rheumatic fever also is associated with widespread collagenous degeneration and vascular changes. There may be many areas of perivascular exudation which form nodules during healing. The lungs are involved in about half the patients during the acute stages and later roentgenograms show a diffuse, hazy shadow over the middle and upper portions of the lung fields, more dense peripherally. If heart failure and pulmonary congestion are present, these findings are obscured. Periarteritis nodosa is a focal arteritis with hyaline degeneration, nodule formation, and obliteration of the lumen. Most patients with this disease have pulmonary involvement at some time and may have pulmonary failure in the acute stages. This is associated with transient but massive, symmetrical shadows extending from the hilum toward the periphery so that a "corona" appears about the mediastinum. In the chronic stages small, hazy shadows scattered throughout the peripheral lung fields and accentuated bronchovascular markings at the bases suggest bronchopneumonia. Serial films show migrations of these shadows similar to those described in Loeffler's syndrome. Exfoliative dermatitis may be accompanied by roentgenographic evidence of a diffuse inflammatory process, disappearing and reappearing with the changes in the skin. Acute glomerulonephritis may alter the permeability of the vascular walls and produce evidence of pulmonary edema and pleural effusion.—*The roentgen appearance of the chest in diseases affecting the peripheral vascular system of the lungs, R. P. Barden & D. Cooper, Radiology, July, 1948, 51: 44.—(G. F. Mitchell)*

Surgery of Tension Cysts.—Congenital cysts of the lung are uncommon. A survey in 1933 included only 150 cases, very few of which were seen in infants or diagnosed during life. The large single tension type of cyst is even more rare. It is bronchogenic in origin and until recently was fatal. In the past five years a few resections have been reported and the authors successfully removed a giant cyst in an infant of four months. The patient's mother had had four previous pregnancies, two of which had resulted in miscarriages and one in a stillbirth; the other infant died during the first day of life. The

patient breathed rapidly from birth but became acutely dyspneic only two days before hospitalization. A roentgenogram of the chest showed an air-filled sac occupying most of the right hemithorax, bulging to the left of the midline, and markedly displacing mediastinal structures to the left. Preliminary decompression by needle and catheter, as suggested by Gross, was done. This procedure gives rise to complications but it allows a return towards normal anatomy and physiology, and aids in controlling infection. After decompression, most of the cyst wall, which was in the middle lobe, was removed at operation. The right upper and lower lobes were expanded at the conclusion of surgery, and the postoperative course and result were good.—*Tension type of congenital pulmonary cyst, J. M. Walker, W. B. Taggart & H. J. Staton, J. Pediat., November, 1948, 33: 601.—(W. H. Oatway Jr.)*

Surgery for Tracheal Compression.—A new method is described for the relief of compression of the trachea by an anomalous artery. An infant had signs and symptoms of tracheal stenosis from birth. For four months the breathing was noisy and wheezing, dyspnea without cyanosis was present, but oxygen was often needed. Rhonchi were heard throughout the chest; leukocytosis and fever were often present. The child insisted on lying in a posture of opisthotonos with the head held in hyperextension. Iodized oil in the trachea showed a defect just above the level of the aortic arch. At operation the right innominate artery was found to arise slightly to the left of the normal position in a common origin with the left common carotid and to press on the right anterolateral surface of the trachea. Rather than interrupt the artery, the surgeon dissected it free from the trachea and sutured it to the periosteum of the posterior surface of the manubrium sterni. The convalescence was satisfactory, the symptoms and signs clearing at once, and a tracheogram showed only a slight residual defect.—*Compression of the trachea by an anomalous innominate artery: An operation for its relief, R. E. Gross & E. B. C. Neuhouser, Am. J. Dis. Child., April, 1948, 75: 570.—(W. H. Oatway, Jr.)*

Multiple Port Irradiation of Deep-seated Tumors.—Roentgenotherapy of deep-seated, malignant tumors, especially of the esophagus and lungs, has been unsuccessful, in part because of the difficulty of delivering a large enough dose to the tumor without damaging the overlying tissues. In recent years, this difficulty has been avoided by several methods including one in which the patient is rotated with the tumor as the axis of rotation during irradiation. The authors have modified this rotation therapy by irradiating multiple fixed fields with the beam so directed that it always passes through the tumor. By this means intensive irradiation is delivered to the tumor while each skin area receives only a fraction of the total dose. The exact extent and site of the tumor must be known, of course, in planning the treatment. Twelve narrow portals of entry on the back and chest and in the axillae are used for tumors of the esophagus or hilar region. Fewer portals are used for pulmonary neoplasms which are more eccentrically located. Carcinoma of the esophagus is especially suited to this form of treatment but solitary tumors of the hilar region and of the lung parenchyma also respond. Gratifying results have been obtained with relief of symptoms, prolongation of life, and reduction in the size of the tumor.—*Treatment of deep-seated malignant tumors with multiple port technic simulating rotation therapy, I. I. Kaplan & S. I. Etkin, Radiology, August, 1948, 51: 188.—(G. F. Mitchell)*

Promizole for Military Tuberculosis.—In general, tuberculosis in adults does not respond well to promizole. However, it was decided to employ this drug at Bellevue Hospital in New York for the treatment of certain forms of tuberculosis in children. It was first used in 5 successive cases of meningitis with no observable beneficial effects.

Acute generalized hematogenous tuberculosis was then selected for treatment with promizole because the course of this disease is more prolonged than that of meningitis, thus allowing more time for the action of the drug. The prognosis of this form of tuberculosis in children is nevertheless uniformly bad. The first cases were started on 1 Gm. daily and the dose was increased rapidly to the point of toxicity. Later the dosage was more slowly increased until a daily dosage of 5 Gm. was reached. A blood level of 2 to 3 mg. per 100 cc. was found satisfactory and, after prolonged treatment, adequate blood levels could be obtained with as little as 1 Gm. a day of the drug. Eleven children, ranging in age from six months to eleven years, with acute miliary tuberculosis received promizole. Treatment was given for one month to two or more years. The toxic effects noted were vomiting, leucopenia, cyanosis, jaundice, a definite increase in the size of the thyroid gland and sex maturation as evidenced by enlargement of the nipples and breast tissue and growth of pubic hair. These changes yielded readily to treatment except possibly the sex maturation. Five consecutive cases of acute generalized miliary tuberculosis were apparently adequately treated with promizole. Of these 2 children died forty-three days and four and a half months after therapy was begun. Roentgenographic evidence of miliary tuberculosis disappeared completely in 3 children who are alive thirty to thirty-three months after promizole was first given. The primary pulmonary lesions cleared slowly and in 2 cases cavitation appeared. One child developed tuberculosis of the spine, choroiditis and positive urine cultures while taking the drug. All three of these cases are still receiving maintenance doses of promizole. Thus promizole may have a favorable action in suitable cases of miliary tuberculosis. It can be given safely and effectively over a period of years.—*The treatment of miliary tuberculosis with promizole. E. M. Lincoln, S. Stone & O. R. Hoffman, Bull. Johns Hopkins Hosp., January, 1948, 82: 1.* (*J. S. Woolley*)

Streptomycin for Tuberculous Meningitis. Thirty children with tuberculous meningitis between the ages of 9 months and 11 years were treated with streptomycin. The drug was administered intramuscularly as well as intrathecally to 25 patients; 5 others were treated with intrathecal injections alone. A series of 20 to 25 intrathecal injections was given, each dose being 0.025 to 0.1 Gm. The intramuscular dosage was 0.25 to 0.5 Gm. daily and was continued for three to five months. Among these 30 cases there were 11 deaths; 19 children were alive after two to fifteen months. The best results were obtained in those children in whom treatment was started not later than on the tenth day of illness. The spinal fluid became completely normal in only 6 of the surviving 19 cases.—*Streptomycin in tuberculous meningitis in children (Russian), N. O. Vassilevich, Probl. Tuberk., 1948, 5: 22.—V. Leites*)

Streptomycin in Chest Surgery.—Reports of the use of streptomycin in pulmonary and pleuropulmonary infections are still scanty. A series of 17 cases of lung abscess, nontuberculous empyema, and tuberculous empyema is reported as a part of the experience with streptomycin in U. S. Army Hospitals. In 3 cases of lung abscess the drug in conjunction with penicillin seemed helpful when the organisms were sensitive but not when the lesions were chronic and extensive. Five cases of nontuberculous empyema were helped remarkably, especially when susceptible organisms, notably *E. coli*, were present and adequate surgical drainage was obtained. In 9 cases of tuberculous empyema the results were variable. Fistulas and empyemas did not seem to improve if the associated pulmonary lesions were productive or if they did not improve coincidentally. It is believed that streptomycin may be more effective in conjunction with penicillin; it may control pyogenic invaders during local or intramuscular administration; and it may extend the scope of plastic operations by providing additional protection.—*Streptomycin in surgical*

infections: VI. Lung abscess and empyema, E. J. Pulaski & T. T. White, Ann. Surg., August, 1948, 128: 312.—(W. H. Oatway, Jr.)

Treatment of Pneumococcal Pneumonia.—The authors set out to determine whether it was necessary to maintain appreciable blood concentrations of penicillin in the treatment of pneumococcal pneumonia. Experimental evidence had indicated that it was not necessary. Seventy-nine patients were given 300,000 units of aqueous crystalline penicillin G every twelve hours. In previous years 69 patients had received aqueous penicillin every three hours, while 58 had received penicillin in oil and beeswax. The results of the newer method were just as good as the others.—*Pneumococcal pneumonia treated with aqueous penicillin at twelve hour intervals, P. A. Tumulty & G. Zubrod, New Engand J. Med., December 30, 1948, 239: 1033.—(A. G. Cohen)*

Penicillin for Lung Abscess.—Sixty cases of lung abscess were treated with endobronchial instillations of penicillin immediately after discovery. Healing of the abscess was considered to have been achieved under the following conditions: (1) disappearance of symptoms and of roentgenographic abnormalities with normal bronchograms for a period of at least six months; or (2) disappearance of the abscess cavity on the roentgenogram with some remaining density or slight sequelae on bronchography such as the demonstration of small isolated bronchial dilatations, dislocated bronchi, or a small single cavity. Each patient received an average of 8 instillations through a bronchial catheter. Of the 60 patients treated with this method, 30 had a follow-up period sufficiently long to be evaluated by the above criteria of cure. Fifty per cent of these patients had no residual roentgenographic abnormalities. Twenty-five per cent were considered healed but had residual changes, often demonstrable only by bronchography. There were 15 per cent failures with multiple recurrences. Ten per cent died of other causes or could not be followed. The patients who do not respond to this treatment must be considered candidates for excisional surgery which should not be delayed too long.—*Quelques problemes posés par la penicilline endo-bronchique dans le traitement des abcès du poumon, H. Métras, M. Grégoire, I. Lieutier, C. Gaillard, Presse méd., July 8, 1948: 471.—(V. Leites)*

Routine Roentgenography.—The results of annual roentgenography of the employees of large banking and commercial enterprises in Paris are reported. In a group of 6,000 employees between the ages of 16 and 60, annual roentgenographic surveys were done from 1942 until 1948. The authors compare the results of the case finding program of regular annual surveys with case finding due to other activities of the medical department of the enterprise ("occasional case finding"). The latter activities were: (1) preemployment examinations, (2) chest examination at the time of intercurrent illness or in the presence of suspicious subjective symptoms, (3) chest examination occasioned by the conversion of the tuberculin reaction. Tuberculin testing was routinely carried out on all employees below the age of 25 and repeated every three months as long as there was a negative reaction. In 1942 when "occasional case finding" was not yet well organized, annual roentgenography detected 4 cases of active pulmonary tuberculosis among 2,880 employees (0.14 per cent). In 1943 one case was found among 3,022 persons. In 1945 and 1946 no cases were discovered. In 1947 there was one case among 4,500 and in 1948 one case among 5,938 persons. Thus, from 1943 until 1948 only 3 cases of active tuberculosis were discovered by annual roentgenographic surveys. During this same period more than 30 cases of active pulmonary tuberculosis were found in this employee group by means other than the annual examination, 24 by the medical department itself and 6 by the family physician or the district tuberculosis clinic. Another survey conducted in 1948 discovered among 5,840 bank employees only one case (10.02 per cent) of active pulmonary tubercu-

losis which was unknown to the medical department. In view of these meagre results the value of annual roentgenographic surveys is considered absolutely out of proportion to the cost in stable and homogeneous employee groups supplied with a good medical department. Much more important is the continuous case finding activity of the medical department especially on the occasion of the slightest suspicious symptoms.—*La tuberculose et les examens radiologiques annuals, G. Poix, M. Barièty, & J. Benetaud, Presse med., November 16, 1948: 776.*—(*V. Leites*)

Case Finding in Hospitals.—The routine examination by roentgenography of hospital admissions and outpatients is one of the best methods of finding tuberculous and other lesions of the chest. Although early trials occurred fifteen years ago, no hospital was known to have a routine program in 1938. A survey in 1943 indicated that 56 general hospitals were using the method but recent data on progress have been either incomplete or unusable. The American Hospital Association reported early in 1948 that 879 institutions of all types claimed to be taking "routine chest x-rays." An attempt has been made to determine the practice in general hospitals which comprise 4,539 of the 6,276 registered hospitals but which receive 93 per cent of the 15,829,000 admissions per year. Information was obtained from all of the state health departments, 43 of which now have tuberculosis control officers. It was found that data on the origin of funds and on the type of films were scanty. Data on the cost per patient and the charges per film were usually not computed or correlated. In most hospitals the taking of chest roentgenograms is far from routine. In spite of a lack of definite statistics, there is evidence that the use of routine roentgenography is being widely extended. Several states have very active and widespread plans. Finances are said to be less of a problem to hospitals than administration. At least 247 general hospitals were found to have a program in effect, 27 more have equipment ready to use, and numerous others have plans to initiate a program.—*The current status of routine x-raying in general hospitals of the United States, September, 1948, W. H. Oatway, Jr., Arizona Medicine, January, 1949, 6: 23.*—(*W. H. Oatway Jr.*)

A Nonofficial Agency in Britain.—Last December the National Association for the Prevention of Tuberculosis celebrated its fiftieth anniversary. The Association remains a voluntary body, encouraging research and seeking public support. There is a definite place in the National Health Service for the nonofficial agencies which are interested in the prevention of tuberculosis and the next ten years should see no restriction of its activities. Indeed, the prevention of tuberculosis might well form the main task of medical officers of health who are now freed from the routine of hospital administration. At present the Association is extending its activities to the British colonies.—*Tuberculosis and voluntary action, Editorial, Brit. M. J., December 25, 1948, 4599: 1114.*—(*R. W. Clarke*)

BCG Cultures.—The possibility arose that frequent subculturing might alter the biological properties of BCG strains. The problem could easily be solved if the cultures could be kept under standard conditions. A method used successfully with other organisms is to freeze-dry the cultures. The problem was to determine the efficacy of this method with BCG strains. Various vehicles were tried; serum and gelatin proved best. It was found that the survival rate after freeze-drying was about 66 per cent. The cultures were stored for one to twelve months at both 4°C. and 26°C. The survival rate was not affected by the length of storage. Antigenicity was tested by tuberculin tests on immunized guinea pigs and by protection tests against virulent strains. There was no difference in virulence between the frozen-dried cultures and controls.—*Viability of freeze-dried BCG cultures, J. Unger, Tubercle, January, 1949, 30: 2.*—(*A. G. Cohen*)

EMPYEMA AS A COMPLICATION OF CHRONIC PULMONARY TUBERCULOSIS

OSCAR AUERBACH[1,2]

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Empyema as a frequent complication of chronic pulmonary tuberculosis is in many of its aspects poorly understood. In an effort to clarify some of the controversial points, it was decided to review all of the cases of empyema which came to autopsy. An attempt was made to correlate such factors as treatment with pneumothorax, bronchopleural fistulas, and bacteriologic findings with the development and course of empyema.

COMPOSITION OF SERIES

One thousand eight hundred seventy-four consecutive autopsies of chronic pulmonary tuberculosis were reviewed. Among these there were 311 cases of empyema, an incidence of 16 per cent.

Diagnostic criteria: All cases in which purulent material was present in the pleural cavity were included in this series. No distinction was made in this study between gradations of the pus as to consistency, specific gravity, or bacteriological variants within the fluid.

Age: The youngest person in the series was 8 years of age, while the oldest was 70 years of age. From table 1 it may be seen that the majority of cases occurred in the age period between 20 and 40 years, the age period in which the greatest number of cases of chronic pulmonary tuberculosis is observed at autopsy.

Sex: There were 205 men and 106 women. This was in direct proportion of men to women in the general autopsies during this period.

Color: One hundred and ninety-one (61.09 per cent) were white, 118 (37.9 per cent) were Negroes, while two (0.6 per cent) were of the Asiatic race. This is also directly proportional to the color distribution in the general autopsy series.

Artificial Pneumothorax

Two hundred and fifty-two (81 per cent) of cases of empyema occurred in the presence of an artificial pneumothorax on the same side, while fifty-nine (19 per cent) were found in cases without antecedent treatment.

Duration of chronic pulmonary tuberculosis before induction of artificial pneumothorax: In table 2 it may be seen that 110 cases (42 per cent) in the series were those in which pneumothorax had been started one to three months after the onset of chronic pulmonary tuberculosis. Another 70 occurred in patients in whom pneumothorax had been started between the fourth and seventh months

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[2] This material was gathered while the author was at Sea View Hospital, Staten Island, New York.

after the onset of the disease. In the remaining 72 cases (28 per cent), the pneumothorax was begun from eight months to sixteen years after the onset of the disease.

Duration of artificial pneumothorax: It may be seen in table 3 that the pneumothoraces in the series were maintained from one month to five years before abandonment. Occasionally pneumothorax was abandoned because the collapse was considered as inadequate, the empyema subsequently developing in the patent pleural space. In some instances collapse therapy was interrupted because of clinical signs of a bronchopleural fistula. In the vast majority of cases, however, the reason for the abandonment of the artificial pneumothorax was the appearance of purulent fluid in the pleural cavity.

TABLE 1

Age Distribution

(311 cases of empyema complicating pulmonary tuberculosis)

	Age							
	1 to 9	10 to 19	20 to 29	30 to 39	40 to 49	50 to 59	60 to 69	70
Number of Cases	1	19	116	93	50	25	6	1

TABLE 2

Duration of Chronic Pulmonary Tuberculosis before Induction of Artificial Pneumothorax

Number of Months	Number of Cases	Number of Months	Number of Cases
1	49	26	2
2	30	27	2
3	31	28	2
4	18	29	1
5	20	30	1
6	16	31	1
7	16	32	0
8	3	33	1
9	6	34	2
10	4	35	0
11	8	36	2
12	4	37	0
13	2	38	1
14	5	39	1
15	1	40	0
16	1	41	0
17	1	42	1
18	1	43	0
19	0	44	1
20	1	45	0
21	0	46	0
22	1	47	1
23	0	48	1
24	0	49 to 282	13
25	1		

In 150 cases (59.5 per cent) the pneumothorax was abandoned from one to eight months after the initial induction. In 55 instances (21.8 per cent) collapse therapy was abandoned from nine to eighteen months after it was started. In the remaining 47 cases (18.7 per cent) it was given up from nineteen months to five years later.

When the pneumothorax had been maintained for a long period of time before the development of the empyema, the bronchopleural fistula, when found, was at the base of a chronic cavity in the upper lobe of the lung (figure 4b).

TABLE 3

Duration of Artificial Pneumothorax

NUMBER OF MONTHS	NUMBER OF CASES
1	30
2	23
3	18
4	22
5	15
6	13
7	14
8	15
9	4
10	5
11	9
12	10
13	4
14	8
15	3
16	3
17	3
18	6
19 to 60	47

From this study it seems justifiable to state that in cases of artificial pneumothorax empyema is an early complication of the therapy, but may occur so long as the collapse is maintained in the presence of an open cavity.

PATHOLOGIC ANATOMY

In the vast majority of cases the empyema cavity was located in the lateral aspect of the chest. It usually extended from the inferior aspect of the upper lobe to the base of the lower lobe. The cavity extended for a variable distance toward the anteromedian and also toward the posteromedian aspects of the chest. In all regions the boundary of the empyema cavity was the fused visceral and parietal layers of the pleura. In most instances the empyema cavity measured about 15 cm. in the apico-basal direction and 12 cm. in width. There were variations in the size and position of the empyema space. In some cases the empyema cavity continued for a variable extent along the diaphragmatic surface. Less frequently it extended toward the apical aspect of the lung. In a

few cases the empyema cavity measured from 3 to 4 cm. and was localized over a bronchopleural fistula. Occasionally the empyema cavity was present in the medial aspect, usually associated with a bronchopleural fistula in the right middle lobe.

The amount of pus within the pleural cavity at the time of the autopsy varied greatly (100 cc. to 3 L.) and depended to a great extent upon how much fluid had been withdrawn just prior to death. The pus was thick in some instances; thin in others. The color varied from grey to green and sometimes had a red tinge due to blood.

The wall of the empyema cavity in early cases was thin, measuring less than one mm. in thickness. It was formed by the slightly thickened visceral and parietal pleurae. The pleural surfaces were lusterless and revealed small amounts of attached fibrin. The fluid in the pleural cavity was small in amount, thin, and contained fibrin.

In the more protracted cases the pleurae were thicker, the process being more marked in the parietal layer. The inner surfaces were lined by a thicker layer of fibrin. Beyond this there was a narrow red zone of vascular granulation tissue. These cavities contained larger amounts of pleural fluid which was thicker in consistency (figure 6).

In the oldest cases the pleurae were still thicker and in advanced cases reach a thickness of 2 to 3 cm. in the parietal aspect and one to 2 cm. in the visceral aspect. The increased thickness was due to a progressive increase of the grey-white fibrous zone beyond the red zone of granulation tissue. It is of interest that in the visceral aspect this grey zone was not always an intimate part of the visceral pleura but was attached to the latter by a thin membrane. Thus, in those cases in which there has been a complete closure of a bronchopleural fistula and an anatomic healing of the tuberculous process within the lung, i.e., closure of the cavities and conversion of the sputum to "negative", a pleurectomy is possible and indicated. Re-expansion of the underlying lung is possible if the pulmonary tuberculous process has caused no excessive destruction and fibrosis of the lung.

The thickened parietal pleura was intimately adherent to the overlying endothoracic fascia and the soft tissue of the chest wall. The pleura was so firmly connected with the overlying tissue that it could be removed from the chest cavity only with great difficulty. The greater portion of the parietal pleura was grey-white connective tissue, but in its outer portions fat might be found. Removal of the pleura from the chest wall was usually difficult, since there is no plane of cleavage. The outer surface of the pleura was attached to muscle. The pleura was often of cartilaginous consistency and contained deep grooves underlying the sites of the ribs (figure 4a).

Bronchopleural Fistulas

Bronchopleural fistulas were present in 153 cases in the series (49.1 per cent). In 112 cases there were single openings and 41 cases with multiple fistulas.

Where multiple perforations were present, they were located either in different lobes on the same side or in the same lobe. The size of the fistulas varied from less than one mm. to that of almost an entire lobe; the latter having occurred

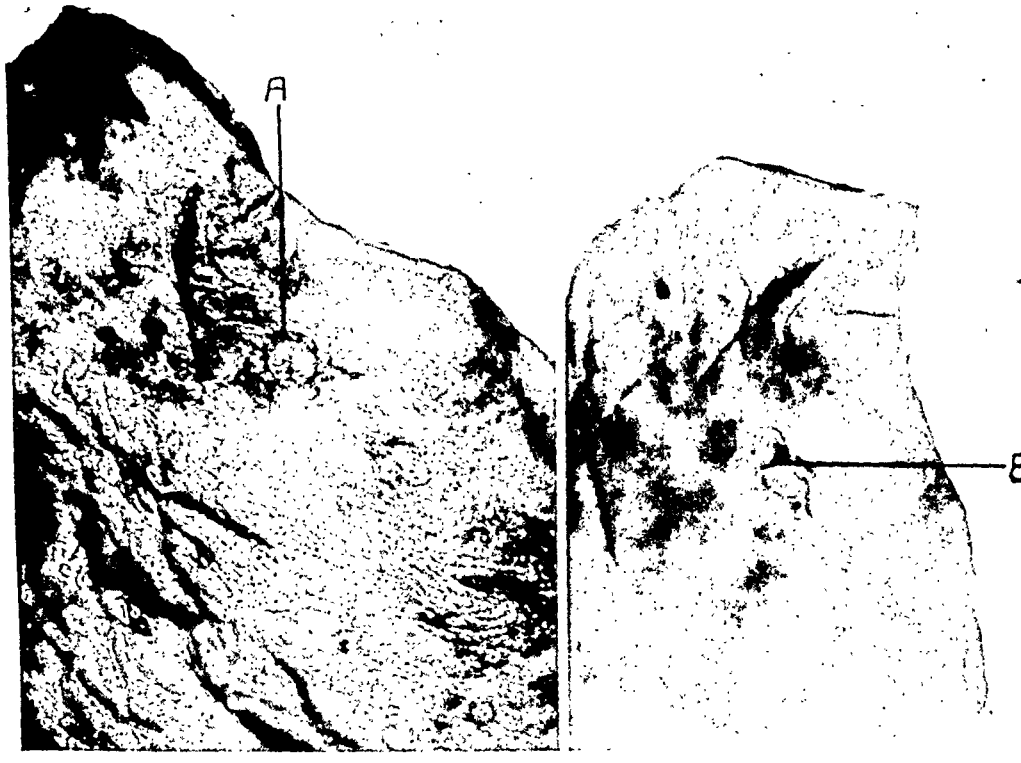

FIG. 1a. (Left) The visceral pleura over the right lung of a 29 year old man who had been receiving artificial pneumothorax on the right side for one month before his death. The total duration of his disease was sixteen months. The patient expired two days after definite clinical signs of a spontaneous pneumothorax on the right side. After diligent search a cap of fibrinous tissue (a) was found firmly attached to the visceral pleura.

FIG. 1b. (Right) Removal of the fibrinous cap in figure 1a (b) revealed a 2 mm. opening in the visceral pleura which communicated with a large cavity in the upper lobe of the right lung.

where artificial pneumothorax was instituted in cases of extensive caseous lobular pneumonia (figures 2a and 2b). The distribution was as follows:

Upper lobe of the left lung	52
Lower lobe of the left lung	19
Both lobes of the left lung	8
Upper lobe of the right lung	44
Middle lobe of the right lung	4
Lower lobe of the right lung	16
Upper and lower lobes of right lung	8
Middle and lower lobes of right lung	2

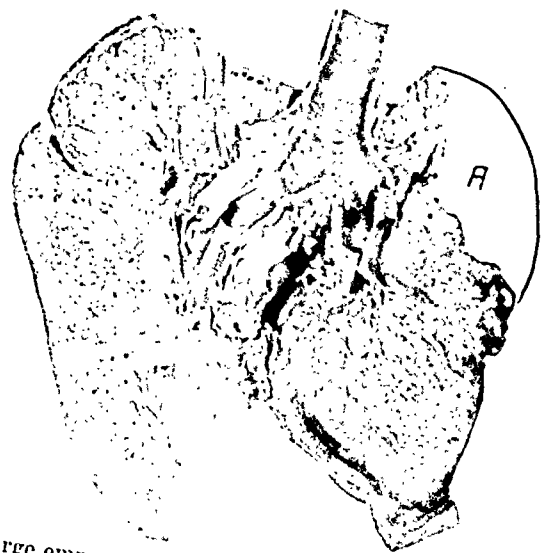

FIG. 2a. (Upper) A large empyema cavity on the left in a 32 year old man whose total duration of illness was seven months. An artificial pneumothorax was begun three and one-half months after the onset of the disease. The parietal pleura is slightly thickened and lined by a grey granular surface. A confluent caseous lobular pneumonia is present throughout both lungs. Tubercle bacilli and staphylococci were found in the pleural fluid during life and *B. pyocyaneous* were recovered after a thoracotomy was performed.

FIG. 2b. (Lower) The same view as figure 2a with the parietal pleura removed. The greater part of the upper lobe of the right lung (a) has been excavated into the pleural cavity. The black line indicates the former border of the lung parenchyma.

Microscopic Appearance

The morphologic appearance of the inflammatory process was a uniform one in all cases, and the same in cases where the bacteriologic studies showed pyogenic organisms or tubercle bacilli as in those in which no organisms were found

in the pleural exudate during life. The presence of a zone of caseation lining the pleural surfaces in the last group would indicate that the pleural fluid had not been sterile.

The inner aspect of the pleura in the early cases was lined in part by a variably wide pink granular zone of caseation and in part by a zone of fibrin containing polymorphonuclear leukocytes and nuclear debris. Toward the periphery of this zone were fibroblasts and epithelioid cells. A narrow to wide zone of vascular granulation tissue surrounded the inner layer. This was composed of fibro-

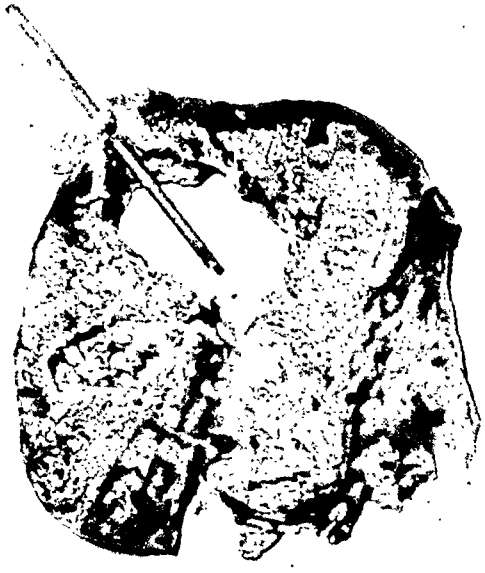

FIG. 3. A coronal section of the upper lobe of the right lung of a 29 year old Negro woman whose total duration of illness was twenty-one months. An artificial pneumothorax on the right side was begun seventeen months after the onset of her disease. A Jacobeus operation to sever an adhesion was performed three weeks before her death. A portion of the severed band is still present, but most of it has sloughed away. A bronchopleural fistula is located at the site of the adhesion band.

blasts, epithelioid cells, Langhans' giant cells, dilated capillaries, and collagen fibrils. In some areas the cellular elements were arranged to form tubercles. Tubercle formation and giant cells were generally not as frequently found as in the tuberculous granulation tissue in the lung parenchyma. It was often necessary to examine multiple sections of the pleura to establish the tuberculous character of the process with certainty.

Beyond the granulation tissue was a zone of loose connective tissue, wider in the parietal than the visceral aspects, which became progressively thicker as the process continued.

Bacteriology

Some classifications (1, 2, 3) of empyema contain two groups and are so divided according to the bacteriologic findings of the fluid. One group is tuberculous

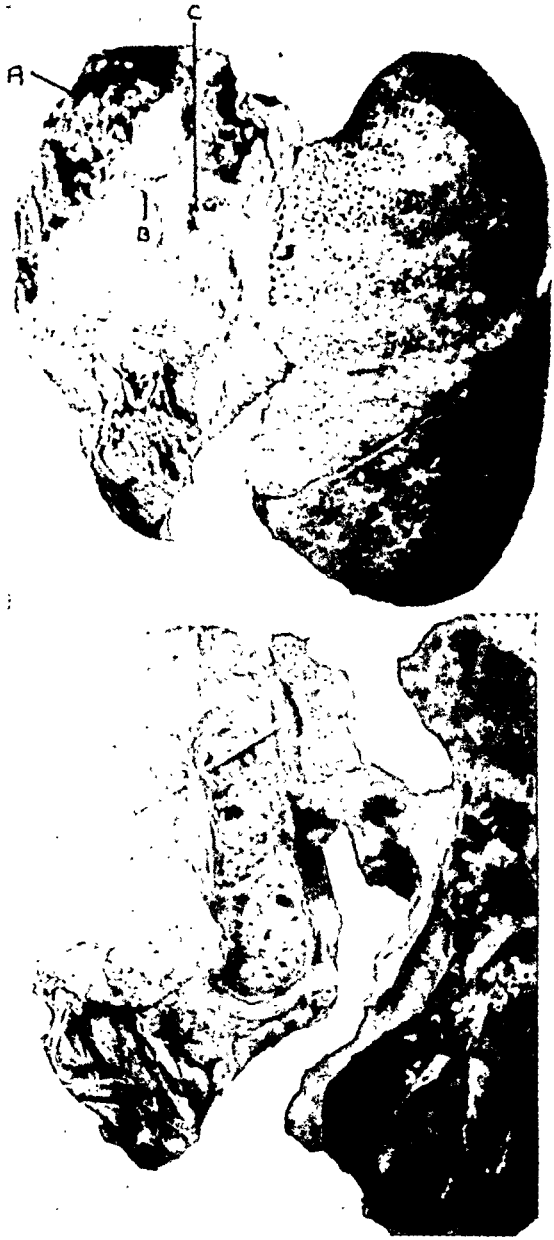

FIG. 4a. (Top) A large empyema cavity on the right in a 26 year old woman whose total duration of illness was forty-four months. An artificial pneumothorax on the right side was begun six months after the onset of her illness and abandoned thirty-one months later because of fluid in the pleural space. The greatly thickened parietal pleura (a) is adherent to the muscles of the chest wall. An adhesion band (b) is present between the visceral and parietal pleurae. The opening of a bronchopleural fistula is seen in the visceral pleura (c).

FIG. 4b. (Bottom) A coronal section of the right lung (figure 4a) shows it to be greatly compressed. A probe is seen passing through the cavity and the opening in the visceral pleura. Only tubercle bacilli were recovered from the pleural fluid during life.

empyema in which only tubercle bacilli are recovered, and the other mixed infection empyema in which in addition pyogenic organisms are also found.

In the present series, bacteriological examination of the pleural fluid was not performed in 101 cases. In the 210 cases in which a study of the fluid was made, 89 showed the presence of tubercle bacilli in pure culture while 78 contained tubercle bacilli and pyogenic organisms. In 19 cases only pyogenic organisms were found and in 24 cases the cultures were sterile.

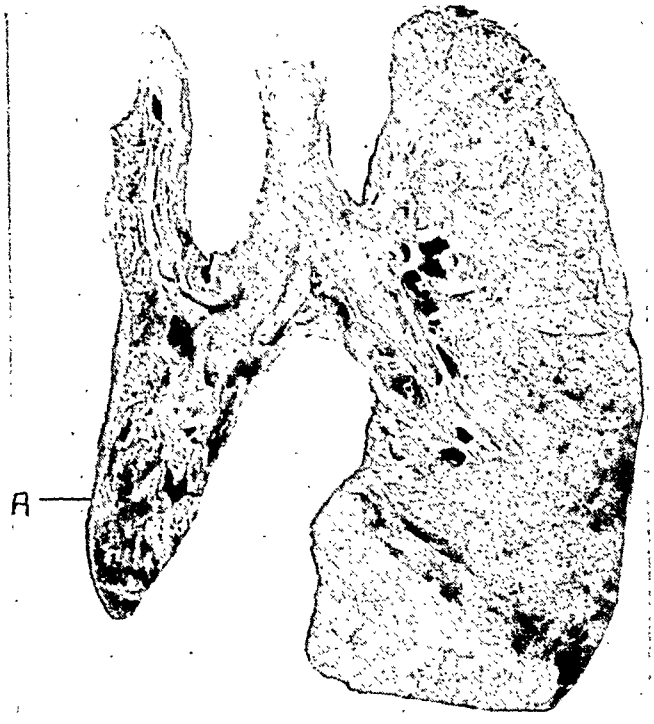

FIG. 5. A greatly compressed lung on the left due to an empyema cavity in a 49 year old man whose total duration of illness was six years. An artificial pneumothorax on the left side was begun two and one-half years after the onset of disease. Death occurred during a second stage thoracoplasty (left) when the pus in the pleural cavity was aspirated into the right lung through a small bronchopleural fistula (a) in the lower lobe of the left lung. The compressed lung is resilient throughout its greater extent. The pleurae over the upper part of the lung are greatly thickened and the pleural space obliterated. Streptococci and tubercle bacilli were cultured from the pus during life.

Except for *M. tuberculosis*, staphylococci were most frequently found in the pleural cavity. This finding is in agreement with the observations reported in the literature. A few authors (4, 5, 6) have pointed out that the finding of staphylococci in an empyema in itself suggests that the condition may be primarily tuberculous. Less frequently isolated were pneumococci and streptococci.

In a small group of cases (35) three other organisms were found in the empyema cavity. These were *B. proteus*, *B. pyocyaneous* and in two instances, *E. coli*. It is interesting that in this group the organisms in all instances appeared after

a communication with the outside. The opening to the outside was the result of a thoracotomy nineteen times, an empyema necessitatis three times, and following a Schede operation, thirteen times.

Another feature of the bacteriologic studies is the great variation in the findings obtained from repeated studies of the purulent fluid removed from the same patient. The variations were great and in many instances extremely inconsistent. In some instances no organisms were recorded, other times tubercle bacilli, and at

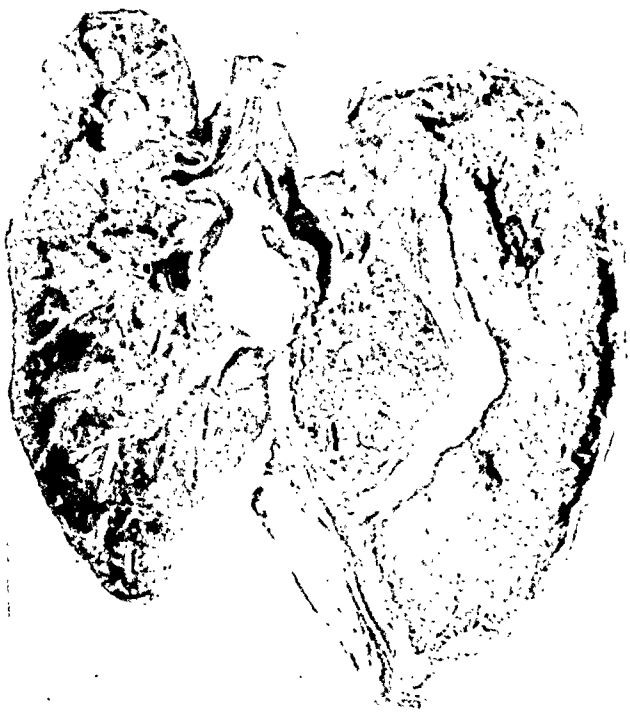

FIG. 6. A large empyema cavity on the left occurring in a 34 year old woman whose total duration of illness was twenty-three months. An artificial pneumothorax on the left side was begun five months after the onset of her disease. A bronchopleural fistula in the upper lobe of the left lung is not visualized in this view. The thickened parietal pleura is lined by a thick green "velvety" surface. An empyema necessitatis which had not ruptured through the skin was present in the left breast. Tubercle bacilli, but no pyogenic organisms, were cultured from the pus removed during life.

other times pyogenic organisms in association with tubercle bacilli. The pyogenic organisms, when recovered, were not always the same.

Healing of Pulmonary Cavities on Side of Empyema

In fifty (16 per cent) of the cases in this series the cavity in the lung had undergone anatomic healing either in the form of an inspissated cavity or a radial scar. In 24 instances cavities were present in the other lobes of the same lung or on the opposite side. In the remaining 26 cases the entire tuberculous process, including the cavity, was either in the stage of healing or was anatomically healed.

In 26 of the 50 cases, an artificial pneumothorax was performed to collapse the cavity and, although in these cases the object was accomplished, the complication resulting from the procedure was the direct or indirect cause of death in many instances. Some of the patients died in uremia secondary to amyloidosis and others of a bronchopneumonia following one of the operations to obliterate the empyema cavity.

Complications

Empyema necessitatis: There were 32 (9.7 per cent) cases in the series which showed evidence of an empyema necessitatis. This is considerably lower than the 24 per cent reported by Sindel (7) in a clinical study from this institution. The basis of this difference lies in the fact that most of the cases in the present series underwent extensive surgical treatment during life in the form of aspirations, thoracostomies, and thoracoplasties.

In all but four instances the empyemata occurred in cases treated with artificial pneumothorax.

In all but one case, the empyema necessitatis was present in the form of an abscess in the soft issue of the chest wall. In the one exception it perforated the esophagus and resulted in a pleuro-esophageal fistula.

The abscess in the chest wall, usually found in the anterolateral chest wall, communicated with the pleural empyema through one or more openings which varied in size from 2 mm. to 2 cm. The latter were enlarged needle tract openings lined by tuberculous granulation tissue. The chest wall abscess varied in size from 2 cm. to 5 cm. in diameter. In 22 of the cases the abscess ruptured through the skin to form a pleurocutaneous fistula.

Amyloidosis: In 107 (34 per cent) of the cases, amyloidosis of varying degrees was present. This compares with 22 per cent in the general autopsy series during this period of time and 35 per cent which were observed in skeletal tuberculosis.

Amyloidosis was present to a much greater extent in those cases in which the empyema had been present for long periods of time. There can be no question that empyema is an important predisposing factor in the development of amyloidosis.

The amyloid deposits were present chiefly in the liver, spleen, kidneys, and adrenal glands. In some of the cases the tuberculous process within the lungs underwent anatomic healing (with cavity closure) and the patient died of uremia, resulting from an extensive amyloidosis in the kidneys.

DISCUSSION

It has been our impression that the cause of the empyema in each instance is a bronchopleural fistula. The importance of bronchopleural fistula as an etiologic factor was recognized by Duboff (8) and stressed by Coryllos (9).

In many of the cases of empyema in the present series, in which there was no bronchopleural fistula present at the time of the autopsy, there was a definite episode which pointed to the presence of one during the patient's life. These episodes included: marked shifts of the mediastinum to the opposite side; the

rapid appearance of highly positive pleural pressures on the affected side; the expectoration of methylene blue when instilled into the pleural cavity; and shortness of breath and pain on the side of involvement.

Bronchopleural fistulas were present in 153 (49.1 per cent) in this series. Woodruff (10) reported an incidence of 30 per cent bronchopleural fistulas in his 154 cases of tuberculous empyema. The reason for his lower incidence lies in the fact that he included in his empyemas any fluid, whether purulent or not, which contained tubercle bacilli. On the other hand, the writer has never observed a case of bronchopleural fistula which had been present for a few days in which an empyema had not been present. It should be stressed that not all cases of spontaneous pneumothorax are followed by empyema. In non-pneumothorax treated cases rapid healing of the fistula with re-expansion of the lung may result in adhesion of the pleural surfaces and obliteration of the space. The writer is in agreement with Leaver and Hardaway (1) that whether or not a patient undergoing pneumothorax therapy develops an empyema depends upon the duration and size of the bronchopleural fistula.

There are cases of bronchopleural fistula which give none of the signs or symptoms just described. These have been designated as "silent", "latent", or "mute" bronchopleural fistulas. Vineberg and Aronowitch (11) believe that the frequent and often undetected presence of bronchopleural fistula in artificial pneumothorax accounts for the increase in empyema which has occurred in the past twenty-five years.

The question arises as to how one may reconcile the presence of so many cases of empyema in the absence of a bronchopleural fistula. The answer to this is that bronchopleural fistulas may heal and in fact do so frequently.

This process is best seen in small fistulas where the healing process is a rapid one. In such cases we have observed a serofibrinous exudate present around the periphery of the recent perforation. This exudate extends to cover the opening and is later transformed into fibrous tissue. At this stage of healing it is difficult and often impossible to find the site of perforation, despite the fact that there had been definite clinical evidence of its presence. Healed perforations cannot be distinguished from subpleural cavities with thickening of the overlying pleura. In a number of cases we were able to demonstrate this method of healing. These patients expired from two to four days after a spontaneous pneumothorax and at autopsy a 2 to 4 mm. fistulous opening was hidden and sealed by a fibrinous cap (figure 1a). Upon removal of the fibrin an open fistula was found (figure 1b). These sites were found with considerable difficulty even in this early stage of healing.

Pollock and Skinner (12) in their clinical observations of cases under artificial pneumothorax found that although the pleuropulmonary fistula is only of a temporary nature it exists sufficiently long to infect the pleural space with tubercle bacilli. Since it was their routine to do fluoroscopic examinations before and after pneumothorax refills, the detection of superimposed spontaneous pneumothorax was often made when unsuspected. Prior to refill they found the

lung compressed in excess to that observed at last fluoroscopy. This finding was soon followed by a pleural exudate.

The emptying of the contents of a pulmonary cavity or liquefied caseous focus into the pleural cavity through the perforation sets up an inflammatory process in the pleura. This inflammatory process, regardless of the type of bacteria involved, has a uniform gross and microscopic appearance. It is not unusual to isolate different species of bacteria from a given empyema cavity at different times.

The finding of pyogenic organisms in the pleural empyema is an extremely interesting feature as the presence of pyogenic organisms in the tuberculous cavity within the lung is a rare occurrence. Yet, in cases with a bronchopleural fistula without thoracotomy drainage, pyogenic organisms are found in the pleural space with great regularity. It is difficult to explain why the bacterial flora present in the bronchi do not gain access to the vomica in the lung, yet when this structure perforates into the pleural cavity, pyogenic organisms are found in the latter. Even in these latter cases, however, pyogenic organisms are rarely found in the pulmonary cavity. A study of the organisms in the empyema cavity (staphylococci, pneumococci, streptococci) indicates that they are the type found normally in the mouth.

The findings of the present study substantiate the often repeated statement that empyema in chronic pulmonary tuberculosis is chiefly (81 per cent of cases) a complication of artificial pneumothorax. It is interesting to note that Vineberg and Aronowitch (11) in their study also found 81 per cent of their empyema cases were secondary to induced pneumothorax. In 2 instances there was empyema present on both sides; both occurred in the presence of a bilateral artificial pneumothorax.

It is unfortunate that in the present series data are not available concerning number of patients with artificial pneumothorax who died during this period so that it would be possible to determine the incidence of empyema in cases of fatal chronic pulmonary tuberculosis with artificial pneumothorax. The incidence as reported by different men (1, 8, 11, 13–16) varies considerably, however, with a range from 5.8 to 40 per cent.

There are three main sources given for the infection: (*1*) rupture of a cavity or a subpleural caseous focus into the patent pleural space; (*2*) tearing of an adhesion; and (*3*) infection from without.

Rupture of a cavity or subpleural focus into the patent pleural spaces: The tuberculous process in the lung usually begins as an area of tuberculous pneumonia in the posterolateral portion of the upper lobe, usually about 2 or 3 cm. above the interlobar fissure. This site corresponds to the infraclavicular shadow observed in the roentgenograms. Surrounding the tuberculous pneumonia is a perifocal reaction the extent of which usually varies with the size of the focus it surrounds. This reaction extends to the pleura and results in a localized serofibrinous exudate. As the process continues the tuberculous pneumonia undergoes caseation, liquefaction, and cavity formation. With the development of caseation, a zone

of vascular granulation tissue develops around it and the perifocal reaction is organized into connective tissue to form the outer wall of the cavity. The perifocal reaction in the pleura is organized into connective tissue so that localized adhesions are formed in the region of the cavity. In later stages this may serve as a protective barrier to the spread of the disease.

If artificial pneumothorax is instituted during the stage of progression, *i.e.*, in the first months of the disease, the protective parietal pleura is then separated from the visceral pleura. When the caseation reaches the visceral pleura and undergoes liquefaction, a bronchopleural fistula develops, the size of which depends upon the extent of the caseous process involving the pleura. In some cases the rupture may be the result of a subpleural caseous focus located in another part of the lobe, or sometimes in the lower lobe.

There is a very important consideration in evaluating the great frequency of empyema in cases where the pneumothorax is instituted soon after the onset of the disease. The question arises as to whether the greater number of cases of empyema at this stage of the disease is due to the fact that the artificial pneumothorax is instituted in the early months of the disease in a greater number of cases than in the later period. Bendove, Miller, and Alexander (17), in an evaluation of 502 cases from the same institution from which the present series is reported, tabulated the duration of the disease before collapse therapy was attempted. They found that the period between the onset of disease and the attempt at pneumothorax was: one to six months in 195 cases; seven to twelve months in 133 cases; and more than a year in 174 cases. Tubercle bacilli disappeared from the sputum in 52.3 per cent of the first group, 39.8 per cent of the second group, and 33.8 per cent of the third group. These writers also noted that the attempt at pneumothorax was unsuccessful in only 12.8 per cent of the first group, in contrast to 25.4 per cent of the third group.

In many cases in the present series the empyema developed some time after the pneumothorax was started. When a bronchopleural fistula was found at the time of autopsy it was present more often at the basal aspect of the unsuccessfully collapsed cavity in the upper lobe. As the tuberculous cavity progresses it extends toward the visceral pleura. When the caseous process which reaches the pleura undergoes liquefaction, it will result in a communication with the pleural cavity at the point where there is no protective wall to cover it. The fistula usually is situated just below the site where both pleurae became joined. Less often perforations were found in the lower lobes, either in the form of a liquefied caseous focus or a cavity rupturing into the pleural space (figure 5).

In the writer's opinion, the great variation in incidence of empyema as a complication of artificial pneumothorax may in part be explained in the choice of cases and the length of time that an unsuccessful pneumothorax is maintained. The more acute the case the greater the opportunity for spontaneous pneumothorax and an empyema to develop. The longer an artificial pneumothorax is maintained in the presence of a persistent patent cavity, the greater is the opportunity for the vomica to rupture into the pleural space with subsequent infection of the latter.

Tearing of an adhesion: Some authors (4, 18–20) believe that in some instances there is a rupture of existing adhesions with infection of the pleura. Joannides (21) believes that the injured pleura comes in contact with contaminated air and this produces an exudate.

The writer has never seen evidence at autopsy of the rupture of an adhesion band with the subsequent development of an empyema. The development of a bronchopleural fistula following the severance of an adhesion has been observed. The latter phenomenon is a very infrequent occurrence, however, and it is believed that the reason for this lies in the structure of the adhesion bands. Three distinct types of bands are present. One is a thin, cord-like band composed entirely of hyalinized connective tissue, another is a broader round band containing pulmonary tissue for varying extent toward the parietal attachment, and the third consists of wide, fan-like adhesions which always contain lung tissue in its parietal attachment. The fan-like adhesions are dangerous to sever and are usually left intact by the surgeon. The great majority of bands are the thin, hyaline connective tissue bands devoid of lung tissue. Section of these bands at their parietal attachment leads to necrosis of the band which halts at its visceral junction. Support for this thesis is found at autopsy since in a great number of cases all traces of the bands severed by previous Jacobeus operation are absent. Lysis of the round adhesion bands containing lung tissue is a probable cause of fistula formation. Under these circumstances, either the coagulative necrosis extends to the pulmonary tissue or the lung tissue is cut directly, leading to a bronchopleural fistula. Since these adhesions are usually found over cavities, the necrosis extends to the lung tissue until the cavity becomes involved. This may come to open directly into the pleural space (figure 3).

Infection from without: In a previous study (22), it was stated that pyogenic organisms can be introduced into a tuberculous empyema from outside the chest wall by means of aspirations, but that this happens so infrequently that its significance is negligible. Leaver and Hardaway (1) state that, although the introduction of pyogenic organisms during pneumothorax refills and during thoracentesis for removal of serofibrinous exudate is given as an etiologic cause, they could find no case in which this was demonstrated. Berry (23) attributed the development of empyema to faulty technique in 3 of 44 empyema cases in which artificial pneumothorax had been administered.

The evolution of a clear serous effusion into a tuberculous empyema has been noted by a number of different observers, among the first of whom were Brauer and Spengler (24) and Dumarest (25). Peters and Woolley (26) state that it is noteworthy that this evolution may take place unnoticed and that it should be clearly differentiated from septic empyema, especially that due to acute lung perforation. The writer believes that in almost all instances the evolution of a serous effusion into an empyema is the result of a bronchopleural fistula which was not discovered clinically. These are the "silent" fistulas to which reference has already been made. This view is shared by Pollock and Skinner (12) who believe that in the infrequent cases in which this occurs there is a spontaneous

pneumothorax superimposed upon an artficial pneumothorax. The pleuro-pulmonary fistula, though of only a temporary nature, exists sufficiently long to infect the pleural space with tubercle bacilli.

Empyema necessitatis: Matson (27) believes that empyema necessitatis occurs as the result of the confluence of many infected puncture sites in the pleura and that, as a result of the destruction of larger or smaller areas of the pleura, the purulent exudate invades the subcutaneous tissue and appears as a bulging mass beneath the skin. Sindel (7) does not believe that the aspiration *per se* plays a role in the empyema necessitatis but that the opening in the inelastic, thick, parietal pleura fails to close rapidly.

Rupture into the lung parenchma: There are some references in the literature to the possibility of rupture of the empyema into the lung parenchyma (27, 28, 29). Jehn (28) states that when a cavity lies in the subpleural region, the pleura is relatively thin, so that it is easy for pleural pus to perforate the cavity wall, with a resulting communication between the pleura, cavity, and bronchi. Matson (27) believes that, when a pleural exudate forms rapidly or exists in the pleural cavity for a long time, the lymph passages undergo an obliterative change, preventing resorption of the exudate. He believes that such exudates usually exist under high pressure and, unless evacuated, tend to rupture into the lung.

The writer's experience is against the generally accepted view that an empyema may extend into the lung parenchyma. In each instance where a pulmonary-pleural communication existed, the cause of the opening could always be traced to the lung either in the form of a cavity or a liquefied caseous focus perforating the pleural membrane.

Healing of the empyema may occur, although it is not a frequent occurrence and can only take place in those cases in which the bronchopleural fistula has healed. The underlying pulmonary tuberculosis in these cases also undergoes anatomic healing. In such instances, the fluid within the pleural space is gradually absorbed. The pleural cavity becomes smaller in size but its lumen does not become obliterated. Both the visceral and parietal pleurae become firmer and progressively more cartilaginous in consistency. The inner surface of the pleural cavity loses its rough granular appearance and becomes smooth.

As emphasized by Hedblom (30), the prospect of re-expansion of the lung is poor after a chronic suppurative pleuritis of the productive type, in which the marked thickening of the pleura is added to the fibrotic changes in the lung.

SUMMARY AND CONCLUSIONS

1. In 1,874 consecutive autopsies of chronic pulmonary tuberculosis there were 311 cases of empyema, an incidence of 16 per cent. Two hundred and fifty (81 per cent) were found in the presence of an artificial pneumothorax and 59 (19 per cent) occurred in cases without antecedent treatment. This substantiates the general opinion that empyema is chiefly a complication of artificial pneumothorax.

2. It is our impression that the cause of the empyema in each instance was a bronchopleural fistula.

3. There was no evidence at autopsy of the rupture of an adhesion band in

artificial pneumothorax (due to stretching) with the subsequent development of an empyema.

4. There is no case in the series in which it could be proved that an infection of the pleural space occurred from without.

5. The evolution of a clear serous effusion into an empyema in almost all instances is the result of a bronchopleural fistula which was not discovered clinically.

6. In 180 cases (72 per cent) the artificial pneumothorax was begun within the first seven months of the onset of the disease. Empyema is generally an early complication of the therapy but may appear at any time as long as the collapse is maintained in the presence of an open cavity.

7. In none of the cases in this series was there evidence of the rupture of an empyema into the lung parenchyma.

8. Empyema necessitatis was a complication in thirty-two (9.7 per cent) of the cases. In all but 4 instances they developed in cases of artificial pneumothorax.

SUMARIO Y CONCLUSIONES

El Empiema como Complicación de la Tuberculosis Pulmonar Crónica

1. Entre 1,874 autopsias consecutivas de tuberculosos pulmonares crónicos hubo 311 casos de empiema, o sea una incidencia de 16 por ciento. En 250 (81 por ciento) había presente un neumotórax terapéutico, y 59 (aproximadamente 19 por ciento) correspondían a casos sin previo tratamiento, lo cual apoya la opinión general de que el empiema es principalmente una complicación del neumotórax artificial.

2. Los AA. tienen la impresión de que la causa del empiema consistía, en cada caso, en una fístula broncopleural.

3. No se observaron signos en la autopsia de la rotura (debida a estiramiento) de alguna adherencia en el neumotórax terapéutico, con la subsiguiente producción de empiema.

4. No hay ningún caso en la serie en que pudiera demostrarse que sobrevino del exterior alguna infección del espacio pleural.

5. La evolución de un derrame seroso límpido en empiema en casi todos los casos representa el resultado de una fístula broncopleural que no se descubrió clínicamente.

6. En 180 casos (72 por ciento) se inició el neumotórax terapéutico en término de los primeros siete meses de la iniciación de la enfermedad. El empiema es generalmente una complicación temprana de la terapéutica, pero puede aparecer en cualquier momento en tanto que se mantenga el colapso en presencia de una caverna abierta.

7. En ninguno de los casos de esta serie había signos de la rotura de un empiema en el parénquima pulmonar.

8. El empiema *necessitatis* representó una complicación en treinta y dos (9.7 por ciento) de los casos. En todos, menos 4, se presentó en casos de neumotórax artificial.

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TUBERCULOUS SPONTANEOUS PNEUMOTHORAX[1, 2]

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INTRODUCTION

Despite the fairly frequent occurrence of spontaneous pneumothorax in pulmonary tuberculosis, relatively little has been written concerning this syndrome. Ornstein and Ulmar (1) in their excellent article quote Biach's finding that of 918 cases of spontaneous pneumothorax, 715 were caused by pulmonary tuberculosis. The present study comprises 40 consecutive patients with pulmonary tuberculosis who developed a spontaneous pneumothorax. On the services from which this report is made the complication of spontaneous pneumothorax occurs in about 1.4 per cent of cases of pulmonary tuberculosis, as 14 cases were detected in 1,000 patients with pulmonary tuberculosis who were admitted consecutively. These 40 patients of the present report were studied as a comparison group with a previously reported series of 63 patients with benign idiopathic spontaneous pneumothorax (nontuberculous) (2).

RESULTS OF ANALYSIS OF SERIES

Symptoms: Symptoms of spontaneous pneumothorax vary from none to a sharp, acute, tearing chest pain with dyspnea, cyanosis, and shock. Of the present group of 40 patients, 30 had sudden pain on the affected side. Since most of these patients were moderately ill with pulmonary tuberculosis, it is possible that their pain thresholds were elevated. Effort was not the cause of the tuberculous spontaneous pneumothorax in any patient. Thirty-one patients had moderate to severe dyspnea, although this was not related necessarily to the degree of collapse of the lung. Thirty-two of the 40 patients had chronic cough at the time of their spontaneous pneumothorax.

Physical signs: Physical examination revealed classical signs of pneumothorax, namely, hyper-resonance, absence of breath sounds, and diminished movement of the affected side of the chest. If pleural fluid was present, flatness was noted at the lung base posteriorly. Examination of the chest roentgenograms revealed that 37 of the 40 patients (92 per cent) had pleural adhesions.

Fluid in pleural cavity: Eleven patients had no free pleural fluid. Six patients had only a small amount of fluid up to the level of the diaphragm, and the remaining 23 patients (57 per cent) had fluid above the level of the diaphragm (table 1). This was in marked contrast to the group of benign idiopathic spontaneous pneumothorax patients, in which group there were only 4 cases out

[1] From the College of Medical Evangelists, Los Angeles, The White Memorial Hospital, Los Angeles, and the Pulmonary Diseases Service, Birmingham Veterans Administration Hospital, Van Nuys, California.

[2] Published with the permission of the Chief Medical Director, Department of Medicine and Surgery, Veterans Administration, who assumes no responsibility for the opinions expressed or conclusions drawn by the authors.

of 85 (5 per cent) with fluid above the level of the diaphragm, and in each of the 4 the fluid was grossly bloody.

Extent of pulmonary tuberculosis: Concerning the degree of tuberculosis, 4 had minimal lesions and 36 had either moderately advanced or far advanced pulmonary tuberculosis. Tuberculous spontaneous pneumothorax may affect either side with equal frequency. In this series, 17 patients had a left pneumothorax, 21 had a right pneumothorax, and two had a right spontaneous pneumothorax first, and later a left pneumothorax. Four patients had subcutaneous emphysema.

Course: The patients who developed tuberculous spontaneous pneumothorax usually were at least moderately ill with their pulmonary disease. The sudden onset of a spontaneous pneumothorax, with or without fluid, added to the patient's respiratory embarrassment and toxemia. Fever and tachycardia were common. Secondary infection of the pleural fluid occurred occasionally, presumably through a bronchopleural fistula produced by the tear of the visceral pleura. The pleural fluid was serous at first, but soon became more cloudy and thicker and of higher specific gravity. Examination of the pleural fluid often revealed acid-fast bacilli (10 of 16 cultures, or 63 per cent). Despite the purulent character of the fluid, however, other pyogens were seldom found (two of 14 patients whose fluid was cultured).

The ages of the 40 patients studied varied from 19 to 70 years. Eleven (28 per cent) of the 40 patients died while in the hospital. Death was caused by both the patient's pulmonary disease and his tuberculous spontaneous pneumothorax. The ultimate mortality rate was probably higher. This cannot be definitely stated since most of these patients were transferred to other tuberculous sanatoriums for further prolonged care.

DISCUSSION

Pathogensis: Spontaneous pneumothorax in pulmonary tuberculosis is usually secondary to subpleural caseation with erosion and rupture of the visceral pleura. The site of rupture may be anywhere along the visceral pleura, probably most often in the upper one-third, particularly the anterior and lateral aspect. Air enters the pleural space and the lung on that side collapses.

According to the literature, subcutaneous emphysema is apparently an uncommon finding. Bloomberg and LaTowsky (3) reported a case of a 41 year old male with pulmonary tuberculosis who had a spontaneous pneumothorax, with subcutaneous emphysema and cerebral air embolism. Hurrell (4) reported a 26 year old male with extensive bilateral pulmonary tuberculosis who developed a left spontaneous hemopneumothorax with subcutaneous emphysema of the tissues of the upper half of the chest.

As 4 patients in the present series had subcutaneous emphysema, this complication is apparently more common than is supposed. A possible mechanism is a sudden tear of the parietal pleura as the result of the rush of air into the pleural space and sudden stretching of an adhesion anchored to the parietal pleura. If of sufficient force to tear the parietal pleura, the free air in the

pleural space dissects through the parietal pleural tear into the thoracic wall and subcutaneous tissue.

Diagnosis: The diagnosis of spontaneous pneumothorax is simple if borne in mind. Sudden chest pain, often with dyspnea, plus the physical findings of pneumothorax as previously noted, indicate the correct diagnosis. The chest roentgenogram is helpful in verifying the degree of collapse, presence of pleural adhesions, and the presence and amount of pleural fluid. If the tear of the visceral pleura is slight, the amount of free air in the pleural space may be small and the patient may have no symptoms. In such instances the diagnosis of

TABLE 1

Contrast of Tuberculous Spontaneous Pneumothorax and Benign Idiopathic Spontaneous Pneumothorax

	TUBERCULOUS SPONTANEOUS PNEUMOTHORAX (40 CASES)	BENIGN IDIOPATHIC SPONTANEOUS PNEUMOTHORAX (85 CASES)*
Pulmonary infiltration on chest roentgenogram	100 per cent	0 per cent
Pleural adhesions	92 per cent	0 per cent
Pleural fluid above level of diaphragm	57 per cent	5 per cent (4 patients had grossly bloody fluid).
Immediate hospital mortality rate	28 per cent	0 per cent
Clinically ill	Almost always.	Not usually, and only very briefly (1 to 2 days).
Fever present	Commonly, usually prolonged, and level may be higher (above 100°).	If present, for only a few days, never more than seven days, and low grade (99° to 100°).
After-care required	Continued bed rest and treatment of pulmonary tuberculosis.	None. Patient can return to full activity after collapsed lung has re-expanded.

* Am. J. M. Sc., April 1948, *215*, 427.

spontaneous pneumothorax can be made only by means of a routine chest roentgenogram.

Contrast with benign spontaneous pneumothorax: This study revealed findings in sharp contrast to a previous review of 85 cases of benign idiopathic spontaneous pneumothorax. In the latter group, pleural adhesions were not demonstrable on the chest film, only 5 per cent had fluid above the diaphragm (grossly bloody in each case), uncomplicated expansion was the rule, the mortality rate was zero, and the chest roentgenogram revealed no evidence of pulmonary infiltration. In general, the patient with benign spontaneous pneumothorax was not ill after the initial chest pain and dyspnea. Fever, if present at all, lasted only one or two days. After re-expansion, the patient with benign spontaneous pneumothorax could return to full activity and was completely well.

Treatment: Treatment of spontaneous pneumothorax in the presence of pulmonary tuberculosis depends on the appreciation of the pathological mech-

anisms and resultant symptoms and signs in the individual patient. If the patient is asymptomatic, with no evidence of tension pneumothorax, such as increasing lung collapse, dyspnea, shift of the mediastinum, and no appreciable accumulation of fluid, then he requires no specific therapy. If the visceral pleural tear has been large, or there is a check-valve mechanism, the affected lung will be greatly collapsed and the patient will be dyspneic and cyanotic. In such a situation, enough air should be aspirated from the pleural space to keep the patient comfortable, and this may be repeated as often as necessary. Oxygen should be used freely by tent or BLB mask. If aspirations of air are required very frequently, an indwelling needle inserted through a sterile cork into the pleural space above the level of any pleural fluid, and attached to a water trap, is satisfactory for reducing the increased intrapleural pressure. This indwelling needle should be discontinued as soon as possible (as soon as there is no bubbling of air into the water trap) because of the possibility of infection of the pleural space via the indwelling needle. In most cases, the indwelling needle, when indicated, need remain in the chest wall for twenty-four hours or less.

The management of the complication of pleural fluid varies. Patients with minimal fluid require no thoracentesis. If dyspnea and cyanosis are present, and these are caused primarily by the presence of the pleural fluid, it should be aspirated. Cough is best controlled by codeine by mouth, and by relieving anoxia and tension pneumothorax when they are present. Persistent tuberculous empyema and a bronchopleural fistula which remains patent usually require thoracotomy and later thoracoplasty. As the tuberculosis is active in virtually all of the patients with pulmonary tuberculosis and spontaneous pneumothorax, the use of streptomycin intramuscularly would be indicated. The chemotherapy should be used both for the treatment of the underlying pulmonary disease and for any tuberculous pleuritis present. Mixed infection empyema usually requires thoracotomy with the preoperative and postoperative administration of both streptomycin and penicillin.

In brief, no fixed hard rules of treatment can be drawn up. Each patient is an individual problem. If the pleural space is infected, the responsible organisms should be determined and treatment varied accordingly. If a bronchopleural fistula is present and remains patent, as determined by air samples from the pleural space with oxygen and carbon dioxide determinations, or by the use of methylene blue or oil of peppermint instilled into the pleural space, treatment will vary depending on whether the pleural space is infected and whether a tuberculous or mixed empyema is present. The patient may be so sick and may have such far advanced pulmonary tuberculosis that death occurs rapidly, often within several days.

SUMMARY AND CONCLUSIONS

1. The case records of patients with pulmonary tuberculosis who developed a spontaneous pneumothorax have been reviewed.

2. It was found that the patients were almost always clinically ill with active pulmonary tuberculosis at the time of the pneumothorax.

3. The onset of spontaneous pneumothorax did not seem to be related to effort.

4. Thirty-seven of the 40 patients had pleural adhesions demonstrable on the chest roentgenogram.

5. Twenty-three patients had pleural fluid above the level of the diaphragm.

6. Four patients had subcutaneous emphysema, a finding previously noted only rarely.

7. The immediate hospital case mortality rate was 28 per cent. The total case mortality rate is probably higher as two-thirds of the patients were transferred to other sanatoriums for further care and hence prolonged follow-up observation was not possible.

8. The proper choice of treatment of tuberculous spontaneous pneumothorax depends upon a number of variables, such as the presence or absence of pleural fluid or tension pneumothorax.

SUMARIO Y CONCLUSIONES

Neumotórax Espontáneo Tuberculoso

1. Este estudio abarcó las historias clínicas de tuberculosos pulmonares que manifestaron neumotórax espontáneo.

2. Observóse que los sujetos se hallaban casi siempre clínicamente enfermos con tuberculosis pulmonar activa al presentarse el neumotórax.

3. La iniciación del neumotórax espontáneo no guardaba al parecer relación con el esfuerzo.

4. Treinta y siete de los 40 enfermos tenían adherencias pleurales observables en las radiografías torácicas.

5. Veintitrés enfermos tenían derrames pleurales más arriba del nivel del diafragma.

6. Cuatro enfermos tenían enfisema subcutáneo, hallazgo este raramente observado antes de ahora.

7. La morboletalidad hospitalaria inmediata representó 28 por ciento, pero la total es probablemente mayor, dado que dos terceras partes de los enfermos fueron trasladados a otros sanatorios para asistencia ulterior, lo cual impidió prolongar la observación subsiguiente.

8. La elección del tratamiento más apropiado para el neumotórax espontáneo tuberculoso depende de varios factores, tales como la presencia o ausencia de derrame pleural o de neumotórax de tensión.

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THE USE OF AUREOMYCIN IN PULMONARY TUBERCULOSIS[1]

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INTRODUCTION

The need for an effective therapeutic agent in the treatment of pulmonary tuberculosis has long been recognized. Although streptomycin has proved to be of value, it is not completely satisfactory because of the frequent development of resistant strains of tubercle bacilli. Furthermore, the high incidence of vestibular dysfunction, which occurs in streptomycin-treated patients, is an additional drawback to its use. For these reasons the search has continued for a drug which would have tuberculostatic properties without the toxic properties of streptomycin.

Aureomycin, an antibiotic derived from a mold, *Streptomyces aureofaciens*, appeared to be a drug worthy of trial in the treatment of pulmonary tuberculosis. It is supplied as the yellow crystalline hydrochloride, highly soluble in water, in which it gives an acid solution with a pH of 4.5. Aureomycin is fairly stable in acid solution but deteriorates rapidly at room temperature in alkaline solution. It is much more effective in acid media than in alkaline, the reverse of streptomycin (1). The emergence of resistant organisms has not been encountered during its *in vitro* or *in vivo* use and there is no evidence of cross-resistance with penicillin, polymyxin, or bacitracin. Aureomycin is rapidly absorbed from the gastrointestinal tract, promptly excreted in the urine, and can be given by mouth (2, 3). The drug has already been shown to have a wide spectrum of activity against many bacteria (4, 5, 6, 15, 16), rickettsia (7, 8, 9, 10, 11), and viruses (4, 8, 12, 13, 14). Duggar (17), using a plate-zone technique, found inhibition of the growth of tubercle bacilli in the presence of growing strains of *Streptomyces aureofaciens*.

Because of these properties and the need for chemotherapeutic agents other than streptomycin in pulmonary tuberculosis, 3 patients with active, exudative forms of this disease were intensively treated with aureomycin.

OBSERVATIONS

Three patients with pulmonary tuberculosis, 2 with tuberculous pneumonia and one with a recent definitely dated exudative "spread", which occurred while the patient was on bed rest in the hospital, were treated with aureomycin[2]. Frequent roentgenograms, sputum smears and cultures for tubercle bacilli, erythrocyte sedimentation rates, complete blood counts, and urinalyses were

[1] From the Division of Pulmonary Diseases, Montefiore Hospital, Bronx, N. Y., and The Montefiore Country Sanatorium, Bedford Hills, New York.

[2] Aureomycin hydrochloride ("Duomycin") was supplied by the Lederle Laboratories Division, American Cyanamid Company, Pearl River, New York.

obtained on all the patients. In addition, liver function tests were obtained in one patient at weekly intervals.

Case Reports

Case 1: (E. D.), a 31 year old white female, with a history of a tuberculous infiltrate in the left upper lobe since 1945, was admitted to the Montefiore Country Sanatorium on March 2, 1948. Three months prior to entry she gave birth to a normal living child. Review of her serial roentgenograms revealed slight regressive changes during 1946 and 1947, and her sputum had been negative for tubercle bacilli for nine months. Nevertheless, it was ap-

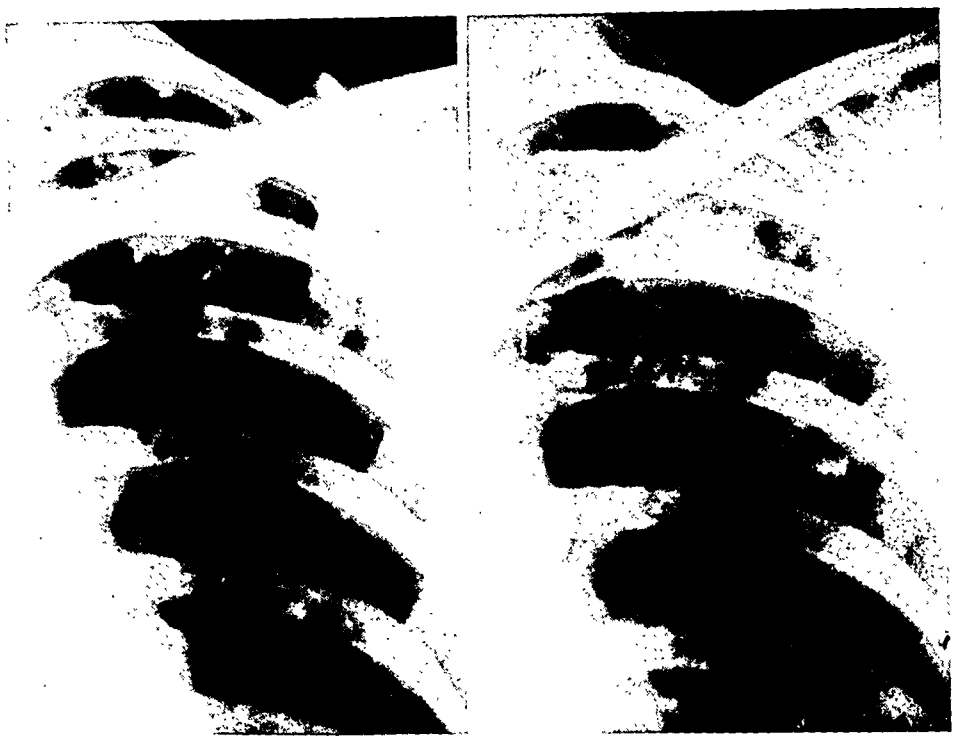
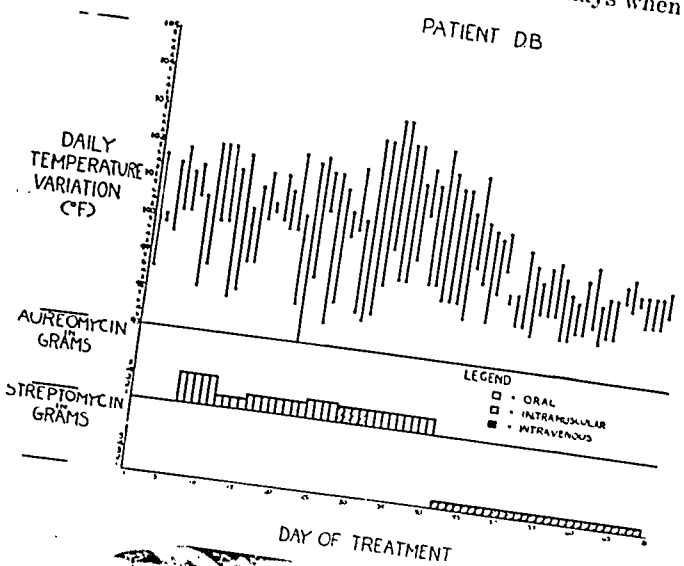


FIG. 1. (E. D.) (Left) September 15, 1948. Recent exudative lesion of the left upper lobe before institution of aureomycin therapy.

FIG. 2. (E. D.) (Right) December 6, 1948. Increase in infiltration in left upper lobe after ten weeks of aureomycin therapy.

parent from her admission roentgenogram that she would probably require active therapy as the lesion appeared "soft" and contained radiolucencies suggesting breakdown. The patient was observed on bed rest for four months without any apparent change on repeated roentgenographic examinations. Her temperature was normal and sputum smears and cultures were negative for tubercle bacilli. In August, 1948 there was an increase in cough and sputum and a wheeze was heard over the left upper lobe. The sputum revealed tubercle bacilli on smears, and roentgenographic examination (figure 1) showed extensive exudative progression of the lesion in the left upper lobe. Bronchoscopic examination revealed acute inflammation at the orifice of the left upper lobe bronchus but no ulceration or granulation. Aureomycin in sixth molar sodium lactate was started intravenously on September 16, 1948. The dose was gradually increased from 300 to 600 mg. twice daily for eight days, at which time the intravenous route of administration was discontinued because of a low grade phlebitis at the sites of injection. Nausea and some drowsiness occurred during intra-

venous administration of the drug. Therapy was instituted by the oral route on the twelfth day of treatment in a dosage of 500 mg. every three hours, a total of 4.0 Gm. a day. This was continued until December 18, 1948 (except for ten days when no drug was avail-



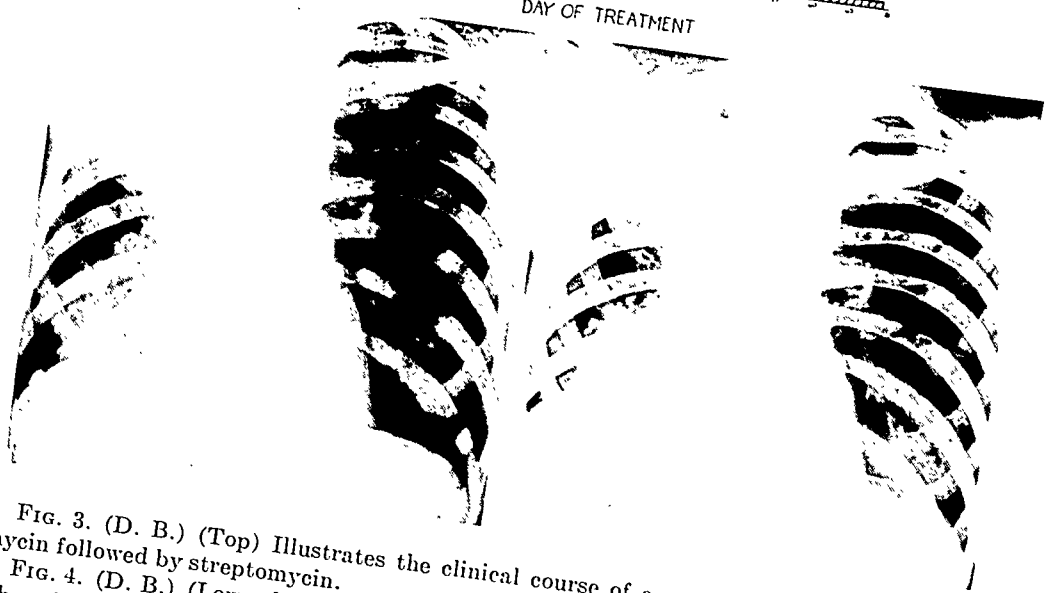


FIG. 3. (D. B.) (Top) Illustrates the clinical course of a patient treated with aureomycin followed by streptomycin.

FIG. 4. (D. B.) (Lower left) October 11, 1948. Tuberculous pneumonia of right upper lobe with bronchogenic spread to right lower lobe. Appearance before institution of aureomycin therapy.

FIG. 5. (D. B.) (Lower right) November 20, 1948. The appearance of the lesion after five weeks of aureomycin therapy.

able) for a total of ninety-four days of treatment. The drowsiness present during intravenous therapy was not noted when the drug was given by mouth. Nausea, vomiting, and anorexia occurred occasionally during oral therapy. Clinical and laboratory observations revealed no alteration in renal or hepatic function nor in the cellular constituents of the

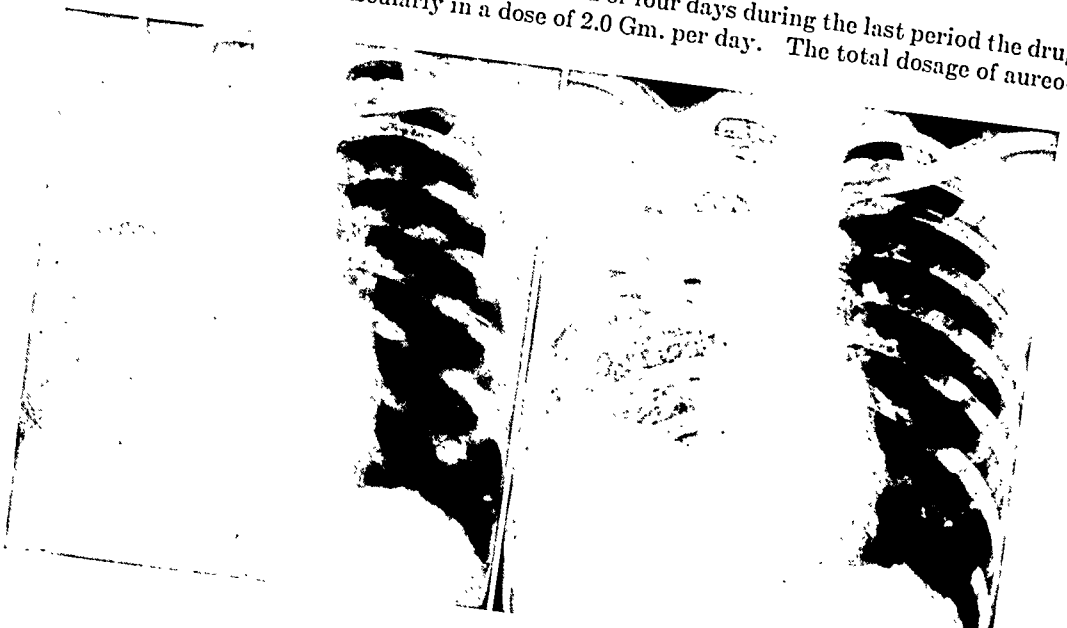
peripheral blood. The temperature was normal throughout the period of therapy. There was some reduction in cough and a decrease in sputum from 15 Gm. to 2 Gm. daily. Nevertheless, the sputum remained positive for tubercle bacilli on direct smear. Serial roentgenograms of the lungs revealed only slight hardening of the peripheral portions of the lesion during the first seventy-five days of treatment. During the third month of therapy a wheeze reappeared over the left upper lobe and the roentgenogram of December 16, 1948 (figure 2) showed an increase in the exudative infiltration in the left upper lobe. Bronchoscopy performed on December 16, 1948 revealed for the first time the presence of granulation tissue at the orifice of the left upper lobe bronchus.

The patient had received a total of 300 Gm. of aureomycin over a period of ninety-four days. Because of the failure to respond to aureomycin and the progression of the disease, the patient was started on a short course of streptomycin preliminary to the induction of a left artificial pneumothorax.

Case 2: (D. B.), a 26 year old Negro female, was admitted to Montefiore Hospital on October 8, 1948 with a history of cough, fever, weakness and weight loss of two months' duration, and a single hemoptysis two weeks prior to admission. This patient's course is illustrated in figure 3. On entry the patient had a temperature of 102° F. Dullness, bronchovesicular breath sounds, and rales were heard on examination over the right upper lobe and at the right base. The sputum was positive for tubercle bacilli on direct smear. The erythrocyte sedimentation rate was 90 mm. in one hour (Westergren method) and the blood leukocyte count was 15,750 with a shift to the left. A chest roentgenogram obtained October 11, 1948 (figure 4) showed an extensive pneumonic lesion of the right upper lobe with bronchogenic spread to the right lower lobe. The patient received aureomycin by mouth every four hours from the seventh to the fortieth day of treatment, in doses ranging from 1.5 to 4.0 Gm. a day. A total of 72 Gm. of aureomycin was employed over a period of thirty-four days. Some nausea, vomiting, and diarrhea were noted when the larger doses were administered and the patient lost several pounds in weight. Slight resorptive changes were noted on the chest roentgenogram (figure 5) but the spiking temperature continued and the sputum remained positive for tubercle bacilli on direct smear. Streptomycin 1.0 Gm. a day was substituted for the aureomycin on the forty-second day of treatment. The effect was promptly apparent in the temperature curve as indicated in figure 3. The temperature returned to normal within several days. Serial roentgenograms showed some clearing of the lesion within fourteen days after streptomycin was started. The patient soon developed a sense of well-being and gained weight. Right artificial pneumothorax was induced on the sixty-third day of treatment.

Case 3: (B. H.), a 30 year old Negro female, was admitted to the Montefiore Hospital on September 13, 1948 with a history of weakness, fever, and weight loss of four weeks' duration. At the time of admission the patient's temperature was elevated to 105°F. and she appeared acutely ill. Signs of consolidation over the entire right lung were elicited on physical examination. The sputum was positive for tubercle bacilli on direct smear; the erythrocyte sedimentation rate was 83 mm. in one hour (Westergren method); there was a moderate microcytic normochromic anemia; and the blood leukocyte count was elevated to 13,450 with a shift to the left. A roentgenogram of the chest (figure 6) showed consolidation of the right upper lobe with an extensive exudative infiltration of the right middle and right lower lobes. Intravenous aureomycin in sixth molar sodium lactate, in gradually increasing doses from 300 to 1,000 mg., was administered from the first to the sixteenth day of treatment. This was discontinued because of a low grade phlebitis at the sites of injection and oral administration of the drug was substituted. During intravenous therapy, drowsiness and some confusion were noted for a brief period. The drowsiness disappeared when intravenous therapy was stopped. The patient then received 4.0 Gm. of aureomycin orally per day in 6 doses from the seventeenth to the forty-third days and 2.0 Gm. a day from the

fifty-fourth to the sixty-fifth days of therapy. For four days during the last period the drug was administered intramuscularly in a dose of 2.0 Gm. per day. The total dosage of aureo-



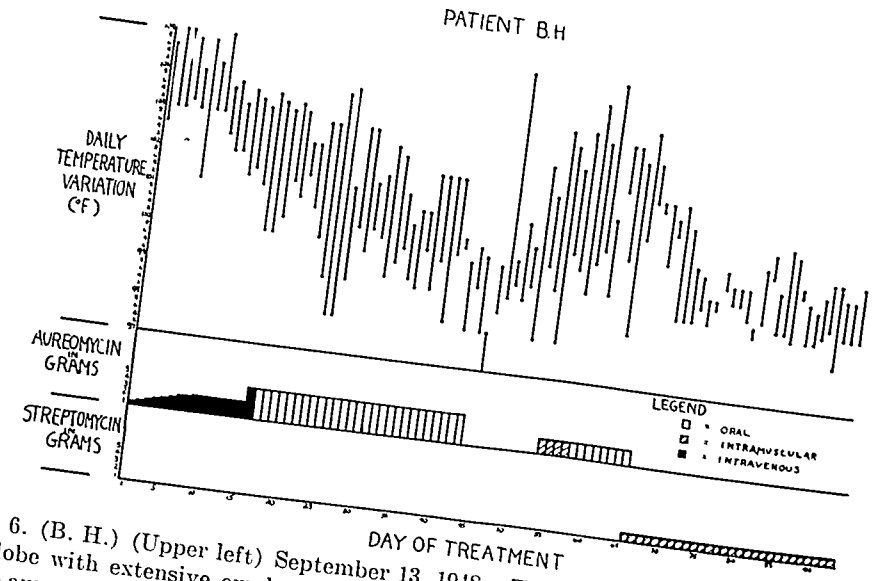


FIG. 6. (B. H.) (Upper left) September 13, 1948. Tuberculous pneumonia of the right upper lobe with extensive exudative involvement of right middle and right lower lobes prior to aureomycin therapy.

FIG. 7. (B. H.) (Upper right) November 20, 1948. Following eight weeks of aureomycin therapy extensive cavitation throughout the right lung is noted. The right leaf of the diaphragm is elevated.

FIG. 8. (B. H.) (Bottom) Illustrates the clinical course of a patient treated with aureomycin followed by streptomycin. Note febrile state responds to streptomycin but not to aureomycin.

mycin was 168 Gm. over a period of sixty-five days. During oral therapy occasional vomiting, diarrhea, a weight loss of 10 pounds, and transitory albuminuria were noted. Cough

and the amount of sputum decreased; the erythrocyte sedimentation rate dropped from 88 mm. to 33 mm. in one hour; and the temperature decreased temporarily but again rose to 105°F. by the time the drug was discontinued. While the patient was receiving aureomycin, progressive excavation occurred in the right lung (figure 7) and the sputum remained positive for tubercle bacilli. During this period spontaneous right diaphragmatic paralysis appeared, probably due to involvement of the phrenic nerve in the thorax by tuberculous mediastinal nodes. Streptomycin 1.0 Gm. per day was substituted for aureomycin on the sixty-sixth day of treatment with prompt decrease in fever and improvement in the patient's general condition (figure 8).

Additional Studies

The tubercle bacilli recovered from the sputum of patient E. D. prior to treatment were tested for *in vitro* sensitivity to aureomycin. Primary isolation of the bacilli on Petragnani's medium was followed by subculture on Dubos' liquid Tween-albumin medium (18). From a diffusely, actively growing 11 day old culture, 0.1 cc. inocula were put into a series of tubes of Dubos' Tween-albumin medium which contained various concentrations of aureomycin. The tubes were incubated for nineteen days. Growth of the organisms was found to be inhibited by concentrations of 10 and 100 γ of aureomycin per cc. No inhibition after the first five days was observed in tubes containing one γ of aureomycin per cc.

Bio-assays[3] of the serum of this same patient for aureomycin revealed the presence of the drug in concentrations up to 8.0 γ per cc. two hours after an intravenous dose of 400 mg. and 1.6 γ three hours after an oral dose of 500 mg. These were the highest concentrations obtained. In patient B. H. (case 3) a concentration of 4.0 γ per cc. of serum was obtained four hours after an oral dose of 700 mg. of aureomycin.

COMMENT

The difficulties in the evaluation of the effectiveness of new therapeutic agents in patients with pulmonary tuberculosis are well known. In order to preclude some of these difficulties in this study, patients were selected with pneumonic involvement, elevated temperature, and evidences of acute illness of apparently recent onset, or with definitely dated recent spread. One patient (D. B.) showed minimal regression radiographically and another (B. H.) showed progressive excavation during aureomycin therapy. In these 2 patients the high fever persisted and the decrease in the severity of the illness was not disproportionate to what might be expected from bed rest alone. The prompt and striking improvement in these patients when streptomycin was used illustrated that the disease could be effectively treated by an antibiotic. In patient E. D. therapy was directed against a new exudative infiltrate and an early bronchial lesion. Progression of both the parenchymal and the bronchial disease was demonstrated during the patient's third month on aureomycin treatment. With streptomycin this is an extremely rare occurrence unless drug-fastness has developed.

The toxic manifestations of aureomycin when given orally for a prolonged

[3] Bio-assays were performed by Mr. A. C. Dornbush of the Lederle Laboratories, using the tube dilution method and a strain of *B. cereus* known as Bacillus No. 5 (3).

period were minimal. Slight nausea, vomiting, and diarrhea were noted. The transitory albuminuria which occurred probably was due to the febrile state of the patient. During intravenous therapy a low grade phlebitis invariably developed and when the drug was given intramuscularly the patients complained of intense pain at the sites of injection.

SUMMARY

Aureomycin was used for periods ranging between thirty-four and ninety-four days in the treatment of 3 young adult patients with acute, extensive, exudative forms of pulmonary tuberculosis. The oral route of administration was more satisfactory than the parenteral route because adequate serum levels were obtained and there was little, if any, toxic effect. Although the drug was found to have tuberculostatic properties *in vitro*, there was no improvement in the patients studied in this investigation. Streptomycin employed as a control therapeutic agent effectively controlled the spread of the tuberculous process.

SUMARIO

Empleo de la Aureomicina en la Tuberculosis Pulmonar

La aureomicina fué empleada en el tratamiento de 3 adultos jóvenes con formas exudativas, extensas y agudas de tuberculosis pulmonar. La droga fué administrada durante períodos de 34 a 94 días. La vía oral de administración resultó más satisfactoria que la parentérica, por obtener adecuados tenores séricos y producir poco, o ningun, efecto tóxico. Aunque la droga mostró propiedades tuberculostáticas *in vitro*, no hubo mejoría en los enfermos estudiados. La estreptomicina, empleada como terapéutica testigo, cohibió eficazmente la difusión del proceso tuberculoso.

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QUESTION OF CONTAGION IN COCCIDIOIDOMYCOSIS. STUDY OF CONTACTS[1,2]

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INTRODUCTION

Recent interest in coccidioidomycosis has resulted from the fact that a large percentage of the hundreds of thousands of military personnel who trained in the Southwest desert area became infected.

It has been shown that the life cycle of the fungus coccidioides immitis consists of two stages: (*1*) a saprophytic stage in the soil found in the desert regions; and (*2*) a parasitic stage in man and animals. It has been believed that the soil (saprophytic) phase could not be by-passed, that man becomes infected by inhalation of the contaminated dust, and that acute infection does not pass from person to person via sputum containing the endospores or spherules (1, 2, 3).

Rosenthal and Routien (4, 5) recently reported, however, that by direct instillation of fungus (spherule) laden material into the respiratory tract of guinea pigs, they were able to infect the lungs of these animals. The material used consisted of undiluted sputum and pus and of emulsions of human granulation tissue and lymph nodes from infected cases. This material was instilled through an opening made in the trachea and was then propelled downward by several injections of air. As a result of these experiments, the authors postulated that spherule-containing material is infectious for guinea pigs when instilled into the lungs. They concluded that active primary or progressive coccidioidomycosis in human beings should be considered contagious until proved otherwise.

The present writers have under observation at this time a group of World War II veterans with residuals of pulmonary coccidioidomycosis. A clinical analysis of these cases has been submitted for publication elsewhere (6). These veterans were all formerly stationed in the Southwest endemic area. All of them returned to their homes in New York City between 1943 and 1946 and have been living with their families from six months to five years. It was felt that, because of the public health aspects involved, it would be important to learn if coccidioidal infection had occurred in the personal family contacts of these cases. To the knowledge of the writers, no study of coccidioidomycosis contacts living outside the endemic area has yet been reported.

METHOD

Contacts of six cases showing a residual pulmonary coccidioidal cavity on chest roentgenograms were investigated. Cases with residual pulmonary nod-

[1] From the Thoracic Unit, New York Regional Office, Veterans Administration.

[2] Published with permission of the Chief Medical Director, Department of Medicine and Surgery, Veterans Administration, who assumes no responsibility for the opinions expressed or conclusions drawn by the authors.

ules or other types of lesions were eliminated. It is felt that the cavity cases would be more likely to infect contacts.

Examinations of the sputum for coccidioidal spherules prior to the return of the veterans to their families were reported negative in 3 of the cavity cases. No record of the sputum findings is obtainable in the remaining 3 cavity cases.

Coccidioidal spherules are not readily recovered from sputum in cases of pulmonary coccidioidomycosis. In 225 cases studied by C. E. Smith[3] the fungus

TABLE 1

Results of Study of Contacts of Cases of Cavitary Coccidioidomycosis

CASES	TYPE OF DISEASE	CONTACT	DURATION OF CONTACT	SKIN TEST OF CONTACT COCCIDIOIDIN 1/100	CHEST ROENTGENOGRAM OF CONTACT
			months		
Case 1	Coccidioidal cavity	Wife—Age 25 Brother—Age 23	30 30	Negative Negative	Negative Negative
Case 2	Coccidioidal cavity	Wife—Age 21	24	Negative	Negative
Case 3	Disseminated coccidioidomycosis (skin, lung)	Wife—Age 27	6	Negative	Not done
Case 4	Coccidioidal cavity	Sister—Age 34 Brother in-law—Age 39 Nephew—Age 3	32 32 32	Negative Negative Negative	Not done Not done Not done
Case 5	Coccidioidal cavity	Wife—Age 29 Son—Age 3 Daughter—Age 4	41 41 41	Negative Negative Negative	Negative Negative Negative
Case 6	Coccidioidal cavity	Wife—Age 23	60	Negative	Negative
Case 7	Coccidioidal cavity	Daughter—Age 2½	24	Negative	Negative

was recovered from only 42 per cent of the cases. These cases were studied in the early stages of infection when the fungus can more readily be demonstrated.

In addition to the cavity cases, a contact to a case of disseminated disease was studied. For obvious reasons, only contacts who had never been in the coccidioidomycosis endemic area were selected for study. All contacts received an intradermal skin test using 0.1 cc. of 1:100 dilution of potent coccidioidin obtained from Dr. C. E. Smith of Stanford University and, where possible, a roentgen film of the chest.

[3] Smith, C. E.: Epidemiology of acute coccidioidomycosis with erythema nodosum ("San Joaquin" or "Valley Fever"), Am. J. Pub. Health, 1940, *30*, 600.

RESULTS

Eleven contacts of 6 cavity cases and one of a case of disseminated disease were examined (table 1). All chest roentgenograms and coccidioidin skin tests of these contacts were negative. If infection had occurred in any of the contacts examined, it would have been expected that a positive coccidioidin skin test would have developed. A positive skin test is known to appear within a week or two after infection and to persist for many years (7).

The results obtained in this study indicate that following a period of six to sixty months of intimate exposure to cases of coccidioidomycosis with pulmonary cavitation none of the contacts had become infected. Although the total number of contacts investigated is not large, it is believed that clinical evidence against the concept of man to man contagion has been shown. Concerning Rosenthal's experiments in the transmission of coccidioidomycosis from man to animals, it should be pointed out that the procedure of direct forceful instillation of infected material into the respiratory tract of guinea pigs goes far beyond the normal relationships obtaining in human beings.

SUMMARY

1. Evidence bearing on the question of contagion in coccidioidomycosis is presented.

2. Eleven contacts of patients with cavitary pulmonary coccidioidomycosis and one contact of a patient with disseminated coccidioidomycosis were studied with chest roentgenograms and coccidioidin skin tests.

3. The roentgenograms and coccidioidin skin tests of all contacts were negative, indicating that none of the contacts had become infected during a period of six to sixty months of exposure.

4. It is believed that additional clinical evidence has been obtained that coccidioidal infection from man to man does not occur.

SUMARIO

El Problema del Contagio en la Coccidioidomicosis: Estudio de los Contactos

1. Preséntanse datos relativos a la cuestión del contagio en la coccidioidomicosis.

2. Once contactos de enfermos con coccidioidomicosis pulmonar cavitaria y un contacto de un enfermo con coccidioidomicosis difusa fueron estudiados con radiografías torácicas y cutirreacciones a la coccidioidina.

3. Las radiografías torácicas y las cutirreacciones a la coccidioidina resultaron negativas en todos los contactos, indicando esto que ninguno de ellos se había infectado durante un período de seis a sesenta meses de exposición.

4. Opinan los AA. que estos datos clínicos aportan neuva prueba de que no ocurre infección coccidioidea de persona a persona.

Addendum

Since this paper was submitted for publication, a reference has been made by Dr. C. E. Smith[4] to several contacts who were studied by him for contagiousness. These contacts were negative to coccidioidin skin testing.

Acknowledgement

The authors wish to acknowledge the assistance of the Bureau of Tuberculosis, Dr. Arthur B. Robins, Director, New York City Department of Health, in performing some of the examinations of the contacts in this study.

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PULMONARY INFILTRATION ASSOCIATED WITH SENSITIVITY TO HISTOPLASMIN[1,2]

Report of a Case

DANIEL W. ZAHN

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INTRODUCTION

As a result of extensive investigation by many workers, particularly those associated with the U. S. Public Health Service, it is becoming increasingly clear that many instances of pulmonary calcification are not the result of previous infection by the tubercle bacillus but, rather, represent the end stage of infection by Histoplasma capsulatum. This suggestion was first propounded by Smith (1) and later elaborated and confirmed by Palmer (2, 4) and by Christie and Peterson (3, 5). Histoplasmin-positive and tuberculin-negative cases of pulmonary calcification were observed to be particularly prevalent in the East Central states. An especially high incidence was noted in the Kansas City area.

Pulmonary calcification associated with histoplasmin sensitivity cannot be differentiated roentgenologically from that which results from tuberculous involvement. This has been emphasized recently by Riley (6). It follows then that the precursors of these calcifications might also show similar features and likewise be indistinguishable. That such is indeed the case has been clearly demonstrated by Furcolow, *et al.* (7). Persistent pulmonary infiltration with histoplasmin sensitivity and tuberculin anergy occurred on 72 occasions among school children in Kansas City, Missouri. Approximately two-thirds of these lesions were nodular and sharply circumscribed. One-fourth were diffuse, patchy, soft lesions which occasionally developed into nodular lesions; a few showed only lymph node involvement, and a few were of the disseminated type. Sontag and Allen (8) also described similar precalcific lesions in Ohio children who were sensitive to histoplasmin but not to tuberculin. It is obvious that the characteristics of these lesions are similar in every way to those of tuberculous infiltrations and that the only means of differentiation at the present time, besides the actual isolation of the offending organism, rests with skin tests. Attempts at isolation of the organism in this type of case have met with little success up to the present time. In only one case was Histoplasma capsulatum recovered (9).

The practical importance of pulmonary lesions associated with histoplasmin sensitivity occurring in individuals who do not react to tuberculin cannot be

[1] From the Tuberculosis Section, Medical Service, Veterans Administration Hospital, Fort Logan, Colorado, and the University of Colorado School of Medicine, Denver, Colorado.

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overemphasized. Such lesions, if not carefully investigated and studied, may lead to an erroneous diagnosis of tuberculosis. Such a case is described below.

CASE REPORT

A twenty six year old white male veteran was admitted to the Tuberculosis Section, Veterans Administration Hospital, Fort Logan, Colorado, on July 24, 1947 with a diagnosis of active minimal pulmonary tuberculosis.

In October, 1946 the patient sought employment with a large corporation in Dayton, Ohio. A routine pre-employment chest film revealed bilateral upper lobe disease, minimal in extent. He was told that he had tuberculosis and that he should seek hospitalization. This advice was ignored, and in June, 1947, the patient prepared to enter a university. Routine stereoscopic photofluorograms again revealed pulmonary disease and hospitalization was advised.

On admission to the hospital, no symptoms referable to the respiratory system were elicited. The past history included pertussis, followed by a moderately severe episode of pneumonia at the age of 8. Scarlet fever, without sequelae, occurred at the age of 23. System review was entirely negative.

The patient's geographical history was as follows: He was born in Springfield, Ohio, where he remained until the age of 12. He lived in Cincinnati, Ohio, from the age of 12 to 24. He lived in Dayton, Ohio, for one year and then entered the Navy in 1943. He spent four months at Great Lakes, Illinois, four months in Gulfport, Mississippi, nine months in Chicago, Illinois, two months in New York City, eight months on duty in the Atlantic and Caribbean waters, one month in San Diego, California, and eight months in the Pacific and Hawaiian waters. He was separated from the Service in Chicago, Illinois, in April, 1946. Since that date, he has spent one year in Dayton, Ohio, and five months in Colorado.

Physical examination revealed the following: height, 5′8½″; average weight, 150 pounds; present weight, 150 pounds; and temperature, 98.6°F. The patient was a well developed and well nourished white male who did not appear to be acutely or chronically ill. He was not dyspneic, cyanotic, or jaundiced. Examination of the head and neck revealed no abnormalities. The pupils were equal and regular and reacted to light and accommodation. The sclerae and conjunctivae were normal. External occular movements were normal. Funduscopic examination revealed no abnormalities. Ear, nose, and throat examination was normal. The gag reflex was present and the teeth were in good repair. Examination of the chest showed a normal thoracic cage. Respiratory movements were equal and symmetrical. The lungs were clear; no rales were noted. The heart was not enlarged. The cardiac rhythm was regular and no murmurs were detected. The pulse was 80 per minute and the blood pressure in mm. of mercury was 120 systolic and 78 diastolic. The abdomen was soft and no masses were palpable. The liver, spleen, and kidneys were not clinically enlarged. The genitals were normal. Rectal examination revealed no abnormalities. The prostate was normal in size, shape, and consistency. Neurological examination was grossly normal. No lymphadenopathy was noted.

Laboratory data were as follows: RBC 5.64 million, hemoglobin 15 Gm., WBC 10,300, neutrophils 57, lymphocytes 39, monocytes 3, eosinophils 1. The erythrocyte sedimentation rate was 4 mm. per hour (Wintrobe). A blood Kahn test for syphilis was negative. Urinalysis was normal.

The following skin tests were performed:

July 30, 1947:

Second strength tuberculin, 0.005 mg. Negative reaction (purified protein derivative)
Coccidioidin, 1:100 Doubtful reaction
Histoplasmin, 1:100 2+ reaction

September 1, 1947:

Old Tuberculin, 0.1 mg. Negative reaction

September 5, 1947:

Old Tuberculin, 1.0 mg. Negative reaction
September 17, 1947:
Second strength tuberculin, 0.005 mg. Negative reaction (purified protein derivative)
Coccidioidin, 1:100 Negative reaction
Histoplasmin, 1:100 2+ reaction
December 15, 1947:
Blastomycin, 1:1,000 Negative reaction

Two specimens of sputum and six gastric washings were negative for tubercle bacilli after eight weeks' culture. Examinations of sputum and gastric washings for Histoplasma capsulatum on smear, culture, and animal inoculation were also negative.

Sternal marrow aspiration was performed on September 22, 1947.[3] Bone marrow smears prepared and stained by Wright and Giemsa stains failed to reveal the presence of the fungus. Culture of the aspirated material was also negative for fungi. Two guinea pigs and 4 white mice were inoculated with specimens of bone marrow. The animals were sacrificed a month following inoculation, and blood smears, as well as smears from the liver and spleen, were negative for Histoplasma capsulatum. Microscopic examination of liver, spleen, lung, kidney, and adrenals revealed no histological evidence of infection by the fungus.

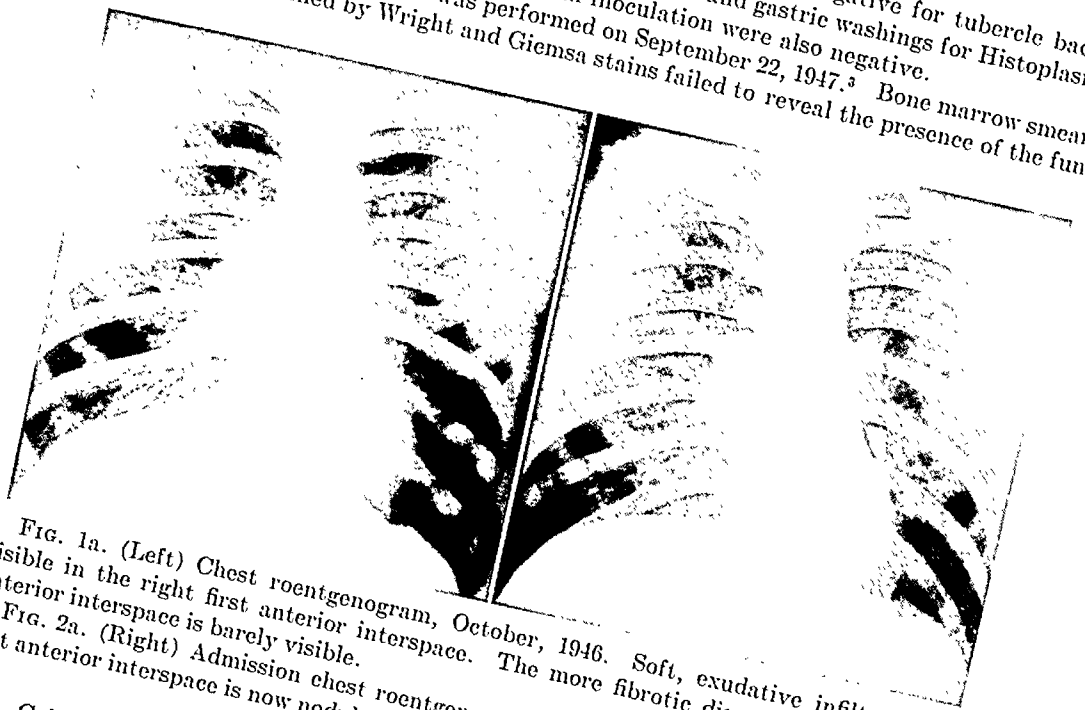

FIG. 1a. (Left) Chest roentgenogram, October, 1946. Soft, exudative infiltration is visible in the right first anterior interspace. The more fibrotic disease in the left first anterior interspace is barely visible.

FIG. 2a. (Right) Admission chest roentgenogram, July, 1947. The lesion in the right first anterior interspace is now nodular and fibrotic in character.

The chest roentgenogram on admission to Fort Logan revealed a minimal productive lesion in the first anterior interspace on the right. A small radiolucent area was present within the density. A smaller fibrotic infiltration was also noted at the periphery of the left first anterior interspace. Apical lordotic films failed to reveal the presence of definite excavation. Fortunately, it was possible to obtain the previous films dated October 18, 1946, taken in Dayton, Ohio. They exhibited a rather soft minimal infiltration in the right first anterior interspace, as well as a more fibrotic lesion in the left first anterior interspace (figures 1a and 1b). A comparison between these films and the admission roentgenograms

[3] These studies were performed by Dr. W. J. Tomlinson, Chief of Laboratory Service, Veterans Administration Hospital, Fort Logan, Colorado.

at Fort Logan revealed that a definite degree of absorption had occurred in the right upper lobe lesion in the nine month interval between the two sets of films. Not only had this density become smaller, but it had also assumed a more productive character. The same was true, to a less extent, of the left-sided lesion (figures 2a and 2b). In view of these changes, it was considered that the lesions certainly were active in October, 1946 and had

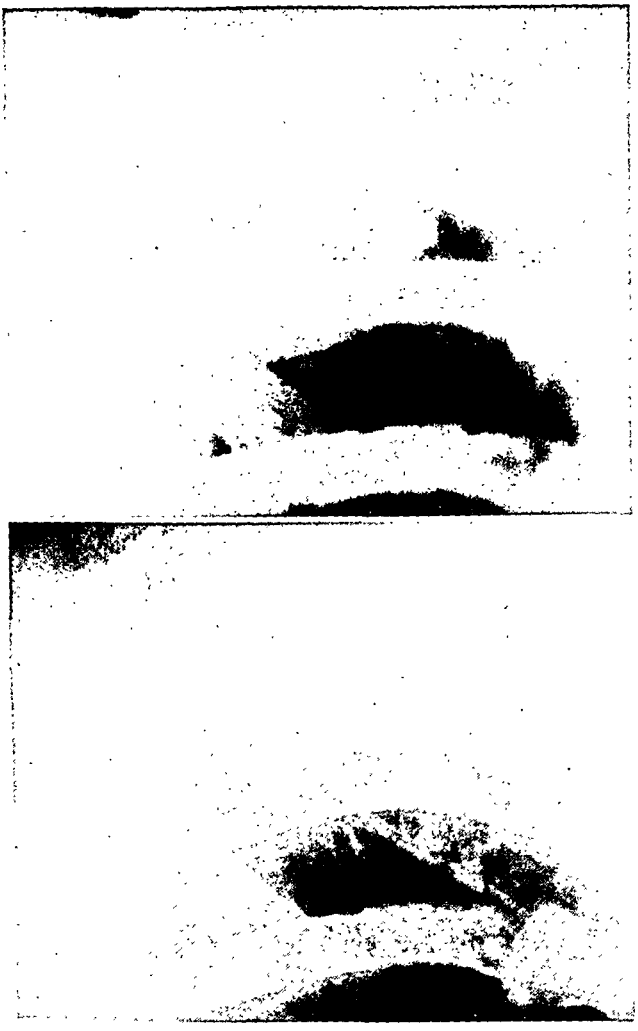

FIG. 1b. (Upper) Enlargement of lesion in right first anterior interspace revealing its soft character.

FIG. 2b. (Lower) Enlargement of lesion in right first anterior interspace. The nodular character of the disease is well visualized.

regressed at the time of admission. Subsequent serial chest films taken at monthly intervals up to December, 1947 have revealed no change in the extent of the disease but the lesion in the right first anterior interspace appears to have become even harder and more fibrotic in character.

The patient's hospital course was entirely uneventful. He was asymptomatic and afebrile throughout. He was placed on strict bed rest on the assumption that his disease was tuberculous in origin. As soon as subsequent studies revealed that such a diagnosis was untenable, however, this rigid regimen was abandoned.

Because of the doubtful coccidioidin skin reaction, coccidioidal serological studies were performed on October 27, 1947. Both the precipitin and complement fixation tests were entirely negative. In November, 1947 histoplasmin and blastomycin serological studies were also performed. The histoplasmin complement fixation test was negative and the blastomycin complement fixation reaction was so slight as to be called negative.

COMMENT

This case meets the three criteria established by Furcolow, *et al.* (7) for possible subclinical cases of histoplasmosis: (*1*) the individual has skin sensitivity to histoplasmin but not to tuberculin; (*2*) the lesion must persist at least two months; and (*3*) laboratory and clinical examinations exclude the presence of tuberculosis and other conditions such as sarcoidosis or Hodgkin's disease.

The failure to isolate tubercle bacilli on repeated attempts, coupled with the persistent tuberculin anergy in the presence of an active lesion (admittedly regressive), make it difficult to label this patient's pulmonary disease as tuberculosis. Houck (10) in an editorial comment emphasized the recent concept that histoplasmosis, instead of being a rare and fatal disease, may be a common and benign infection occurring in a large part of the population of some areas in the Middle West. He stressed the importance of considering the patient's history of residence in endemic areas and the use of appropriate skin tests in the investigation of pulmonary lesions. The reaction to histoplasmin and the long period of residence in what may be termed an endemic area (Ohio) lend support to the belief that this case may represent such a benign type of histoplasmosis as is postulated by Houck.

As noted above, the histoplasmin complement fixation test on this patient's serum was negative. The exact significance of this procedure as a diagnostic measure remains undetermined. The studies of Furcolow and his associates at Kansas City are expected to shed some light on this question. It may be stated with some degree of confidence, however, that negative serological studies do not necessarily eliminate histoplasma as the etiological agent. Some support for this statement may be obtained from the experience with serological studies in coccidioidal infections. In quiescent coccidioidal lesions (with normal erythrocyte sedimentation rates), the complement fixation tests are usually negative (1). If these findings may be transferred to the field of histoplasmotic infections, serological studies in the regressive lesions, as illustrated by this case, would also be expected to yield negative results.

The question of the further management of such a case merits some discussion. There can be little argument with the view that during the period of investigation the case should be treated as one of tuberculosis. The value of strict bed rest in minimal tuberculosis has been adequately demonstrated by Amberson (11), and such a regimen should be immediately instituted. However, when the diagnosis of tuberculosis is shown to be in error by the negative results of skin tests and the failure to obtain tubercle bacilli in sputum and gastric washings by culture and/or guinea pig inoculation, it would appear that such a rigid therapeutic schedule is no longer required. Admittedly, the entire picture of the entity "benign histo-

plasmosis" has not yet been fully observed and, accordingly, definitive statements regarding therapy cannot be made. The supposition that these cases require little or no treatment has no basis in fact, however, and conceivably could lead to serious harm. The tendency following the exclusion of tuberculosis as a diagnosis is to allow the patient unrestricted activity. This conclusion is naturally drawn from the investigations emphasizing the close correlation between histoplasmin sensitivity and pulmonary calcification (2, 3, 4 and 5). The innocuousness of "benign histoplasmosis" has been emphasized by Houck (10). The excellent observations, however, of Sontag and Allen (8) on 170 normal southwestern Ohio children sounds a note of caution. While none of their histoplasmin-positive children who exhibited pulmonary lesions presented symptoms comparable to those seen in cases of proved histoplasmosis, the serial growth data and illness histories of these children strongly suggest that the growth progress and health pattern may be significantly different from the negative histoplasmin reactors. In the light of these observations, it would appear that at the very least these patients should be closely observed and followed with serial clinical laboratory and roentgenographic studies.

SUMMARY

A case of pulmonary infiltration with tuberculin anergy and histoplasmin sensitivity is reported. The possibility of such a case representing an instance of "benign histoplasmosis" is discussed. The danger of confusing such cases with tuberculosis is emphasized and the broad aspects of management of such a case are reviewed.

SUMARIO

Infiltración Pulmonar Asociada a Sensibilidad a la Histoplasmina

Comunícase un caso de infiltración pulmonar con anergia tuberculínica y sensibilidad a la histoplasmina, discutiéndose la posibilidad de que represente un caso de "histoplasmosis benigna." Recálcase el peligro que entraña la confusión de esos casos con tuberculosis y repásase, a grandes trazos, la asistencia de los mismos.

Acknowledgments

The coccidioidin employed in this case was supplied by Dr. Charles E. Smith of Stanford University, California, who also performed the coccidioidal serological studies.

Dr. M. L. Furcolow of the United States Public Health Service, Kansas City, Kansas, performed the histoplasmin and blastomycin complement fixation tests and supplied the blastomycin for skin testing.

The histoplasmin skin reagent was supplied by Dr. J. C. Peterson of Vanderbilt University, Nashville, Tennessee.

It is a pleasure to express my deep appreciation to these workers for their cordial cooperation.

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A CORRELATED STUDY OF TUBERCULIN, HISTOPLASMIN AND COCCIDIOIDIN SENSITIVITIES WITH PULMONARY CALCIFICATIONS IN THE ROCKY MOUNTAIN AREA[1,2]

W. K. ABSHER AND F. CLINE, JR.

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INTRODUCTION

During and since the years of World War II, rather extensive investigative studies have been undertaken concerning the etiologic factors responsible for multiple pulmonary calcifications. The results have cast doubt upon the earlier concept that all multiple lung and hilar calcifications are tuberculous in origin. It now appears that fungus infections may play a significant role in the production of these calcifications and a considerable number of cases have been shown to be associated with histoplasmin skin sensitivity. The chronological development of this subject has been thoroughly reviewed during the past few years by several authors (1, 2, 3, 11, 12) and its repetition seems hardly necessary at this time.

One of the most significant contributions has been that of Sontag and Allen (13) in their study of Ohio school children. These observers noted the presence of soft precalcific infiltrates in histoplasmin reactors and found the characteristics of these infiltrates to be similar in every detail to those of tuberculous lesions.

It follows then that physicians should be keenly cognizant of these observations lest they err in their evaluation of such pulmonary lesions. These implications are especially important today with the mass roentgenographic screening of population groups, projects which will undoubtedly uncover increasing numbers of nontuberculous lesions.

The above studies suggest that a condition such as nonfatal Histoplasma capsulatum infection may exist; however, all investigators stress the fact that this entity occurs only in a relatively small endemic section of the country, namely, the East Central States. Ferebee (5) has recently commented that in some geographic areas such as Colorado it is rare to find a native who reacts to histoplasmin. Therefore, on first thought it would appear that the problem is of no practical concern to the physician of the Rocky Mountain Area. The fallacy of such thinking, however, was quickly and forcibly illustrated to the writers when they were confronted with a case (16) diagnosed roentgenographically as active minimal pulmonary tuberculosis.

Briefly, this patient came from Dayton, Ohio (in the endemic area). His skin reactions were repeatedly negative to purified proteid derivative (PPD), 0.0005

[1] From the Tuberculosis Section, Medical and Radiological Services, Veterans Administration Hospital, Fort Logan, Colorado.

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mg., and the reaction to histoplasmin was positive. Many specimens of sputum and gastric washings were negative for tubercle bacilli by culture and guinea pig inoculation. Obviously, this individual had to be considered as nontuberculous and, in all likelihood, represented a case of subclinical histoplasmosis.

Following this experience, a cursory review of the admissions to Fort Logan revealed the fact that relatively few veterans were natives of Colorado and that a large number had lived most of their lives in the endemic area of the East Central States. A review of the personnel working in the Veterans Administration Regional Office in Denver brought out the same surprising fact. Accordingly, it was decided to conduct a survey of 1,000 veterans and Veterans Administration personnel in the Rocky Mountain Area relative to the problem of pulmonary fungus infections.

METHODS

Each subject was skin tested with purified protein derivative (PPD), 0.0005 mg., coccidioidin in a dilution of 1:100, and histoplasmin in a dilution of 1:100. The results of these skin tests were correlated with a chest roentgenogram in every instance. All skin tests were interpreted by the same group of investigators and all roentgenograms were interpreted by one radiologist who did not know the results of the skin tests. The usual standards were adopted in the interpretation of the skin reactions. In interpreting the roentgenograms, at least 10 calcifications were required before the term "multiple pulmonary calcifications" was applied to an individual case.

RESULTS

Of the 1,000 cases tested, 55.8 per cent reacted to tuberculin, 6.5 per cent reacted to coccidioidin, and 22.6 per cent reacted to histoplasmin (table 1). It will be readily noted that the percentage positive to tuberculin is similar to the findings in other parts of the country. The percentage of reactors to coccidioidin was low, as originally expected. Surprisingly enough, however, the percentage of reactors to histoplasmin was considerably higher than was anticipated, since Palmer (10) in his study obtained only 4 positive reactions in 460 Colorado nurses.

As was expected, many individuals reacted to more than one antigen (table 2). The points of interest here are that 348 (34.8 per cent) of those tested did not react to any antigen and, of this number, only one revealed multiple pulmonary calcifications on roentgenographic examination. Thirty subjects (3 per cent) reacted to all three antigens and, of this group, 5 showed multiple pulmonary calcifications. The largest group was that which reacted to tuberculin alone, and of this group only 14 revealed pulmonary calcifications. One hundred ten, or 11 per cent, reacted to both tuberculin and histoplasmin and in this group 16 showed multiple pulmonary calcifications. Of 73 reactors to histoplasmin alone, 8 showed pulmonary calcifications.

Thus, the tuberculin- and histoplasmin-positive group (T+ H+ C−) showed a higher percentage of pulmonary calcifications than any other group except the

one with positive reactions to all three antigens (T+ H+ C+). In other words, as far as this study is concerned, neither tuberculosis nor histoplasmosis has played a dominant role in the production of pulmonary calcifications.

Of this group of 1,000 cases, only 322 could be classified as native Coloradians; that is, they had spent five-sixths of their lives within the state of Colorado (Palmer's criterion (10) in his original geographic survey). The significant feature is that 40 (12.4 per cent) of these so classified natives reacted to histoplasmin. This is considerably lower than the figure for the group as a whole, which was

TABLE 1

Positive Reactions to Antigens in Survey of 1,000 Subjects

	NUMBER OF POSITIVE REACTORS	PER CENT POSITIVE
Tuberculin (0.0005 mg. PPD)	558	55.8
Histoplasmin 1:100	226	22.6
Coccidioidin 1:100	65	6.5

TABLE 2

Results of Skin Tests in 1,000 Subjects

	NUMBER OF CASES	PER CENT OF CASES
T+ H− C−	404	40.4
T− H− C−	348	34.8
T+ H+ C−	110	11.0
T− H+ C−	73	7.3
T+ H+ C+	30	3.0
T+ H− C+	14	1.4
T− H+ C+	13	1.3
T− H− C+	8	.8

T = Purified Protein Derivative, 0.0005 mg.
H = Histoplasmin, 1:100.
C = Coccidioidin, 1:100.

22.6 per cent, but is much higher than Palmer's (10) original figure of less than 1 per cent for Colorado nurses. A more detailed study of the geographic history of these 40 individuals in the present series, however, revealed that 27 had lived for one or more years within the endemic area of the East Central states. Thus their reaction to histoplasmin could possibly be accounted for by residence in other states and only 13 (4 per cent) remained unexplained on a geographic basis.

DISCUSSION

This roentgenographic and skin testing survey of 1,000 cases has served to confirm fully the previous findings of the U. S. Public Health Service investigators (1, 5, 6, 7, 8, 10), namely, that the Rocky Mountain region is not an endemic area for the so-called benign type of histoplasmosis. Nevertheless, the large

number of migrants to this area, together with those native Coloradans who spent some time in the endemic area of the East Central states during the war years, make the problem an important one to the physician in the Rocky Mountain region. This fact has been adequately demonstrated by the erroneously diagnosed case (16) presented previously.

In their recent article concerning the problem of pulmonary infiltrates associated with histoplasmin sensitivity, Furcolow (7) and his associates in Kansas City implied that a specific type of calcification was characteristic. They referred to a so-called "halo" type of calcification in which a soft areola surrounded

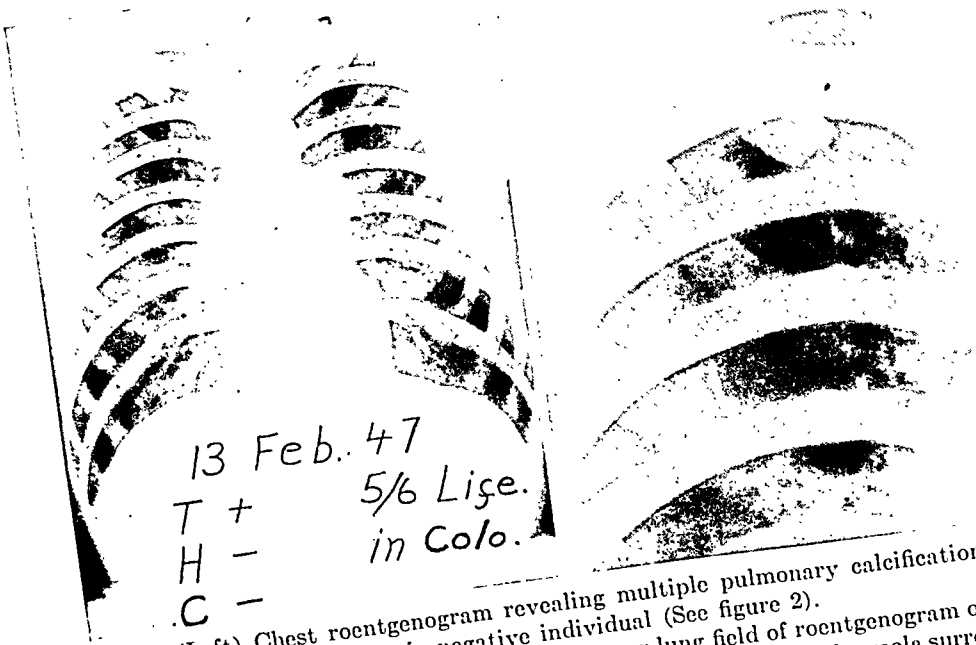

FIG. 1. (Left) Chest roentgenogram revealing multiple pulmonary calcifications in a tuberculin-positive, histoplasmin-negative individual (See figure 2).

FIG. 2. (Right) An enlargement of the right upper lung field of roentgenogram of figure 1 showing the lesion in the first right anterior interspace. Note the soft areola surrounding a central calcific core in this tuberculin-positive, histoplasmin-negative individual.

a central calcific core. That this so-called "halo" calcification is not limited to histoplasmin-sensitive individuals is illustrated in the roentgenograms in figures 1 and 2. The patient whose roentgenograms are presented in the figures exhibited a marked reaction to PPD (0.0005 mg.) and did not react to either histoplasmin or coccidioidin.

Recently Edwards and her associates (4), in reviewing the roentgenographic findings among student nurses, pointed out that calcifications associated with histoplasmin sensitivity were scattered bilaterally, involving both upper and lower lobes. In contrast, tuberculin-positive calcifications usually were limited to the upper lobes. In the present series of cases, however, no diagnostic pattern of pulmonary calcifications was noted in either the tuberculin-positive or the histoplasmin-positive group.

An additional observation in this series was that hilar calcifications, parenchymal calcifications (less than 10), or combinations of the two, occurred in 24 individuals who did not react to any of the three antigens. Long (9) has observed that, of 2,490 cases, 11 per cent lost their sensitivity to tuberculin during a ten year period. Thus there exists the possibility that sensitivity to histoplasmin and coccidioidin may behave in a similar manner. Evaluation of these 24 nonreactors who revealed evidence of previous pulmonary disease is difficult. It would appear to be the consensus of opinion that, from a diagnostic viewpoint at least, a negative tuberculin test rules out the existence of tuberculosis. Woodruff (15) has stated, "A patient anergic to tuberculin, who is not acutely ill, almost certainly does not have clinical tuberculosis." Sweany (14) has remarked, "The absence of tuberculous disease is almost certain in uncomplicated cases negative to 1:1,000 dilution of OT."

Physical examination of these 24 nonreactors revealed no evidence of other diseases, conditions, or metabolic disturbances which, according to Riley (12), are also capable of producing pulmonic calcifications.

With these facts at hand, it would seem that serious consideration should be given to the possibility that there exists yet another unrecognized causative agent or factor capable of producing hilar and/or parenchymal calcifications.

SUMMARY

1. A roentgenographic and skin testing survey of 1,000 cases from the Rocky Mountain Area relative to the problem of multiple pulmonary calcifications has been reported.

2. The survey revealed that 404 (40.4 per cent) of the individuals tested reacted to tuberculin alone and, of these, 14 showed multiple pulmonary calcifications; 73 (7.3 per cent) reacted to histoplasmin alone, of whom 8 showed multiple pulmonary calcifications; 110 (11 per cent) reacted to both histoplasmin and tuberculin, and in this group 16 showed multiple pulmonary calcifications.

3. Twenty-four cases of hilar and/or parenchymal calcifications were noted in individuals reacting negatively to all antigens.

4. Observations in the survey confirm the findings of the U. S. Public Health Service investigators, namely, that the Rocky Mountain Area is not endemic for the so-called benign histoplasmosis.

5. In this series, no diagnostic pattern or distribution of pulmonary calcifications was noted in either tuberculin-positive or histoplasmin-positive individuals.

6. The fact that "halo" type pulmonary calcifications are not limited to histoplasmin-sensitive individuals was noted.

SUMARIO

Estudio Correlativo de las Sensibilidades a la Tuberculina, la Histoplasmina y la Coccidioidina con las Calcificaciones Pulmonares en la Región de las Montañas Rocosas

1. Este estudio radiográfico y cutirreactor comprendió 1,000 personas de la Zona de las Montañas Rocosas con respecto al problema de las calcificaciones pulmonares múltiples.

2. La encuesta reveló que 404 (40.4 por ciento) de los individuos comprobados sólo reaccionaban a la tuberculina, y de ellos, 14 revelaron calcificaciones pulmonares múltiples; 73 (7.3 por ciento) sólo reaccionaron a la histoplasmina, y 8 de ellos mostraron calcificaciones pulmonares múltiples; 110 (11 por ciento) reaccionaron tanto a la histoplasmina como la tuberculina, y 16 de este grupo revelaron dichas calcificaciones.

3. En sujetos negativos a todos los antígenos notáronse 24 casos de calcificaciones hiliares y/o parenquimatosas.

4. Las observaciones realizadas en eta encuesta confirman los hallazgos obtenidos por el Servicio de Sanidad Pública de los E. U. A., o sea, que la región de las Montañas Rocosas no es endémica para la llamada histoplasmosis benigna.

5. En esta serie no se observó ningún patrón o distribución de calcificaciones pulmonares utilizable para diagnóstico, ni en los positivos a la tuberculina ni en los positivos a la histoplasmina.

6. Se notó que las calcificaciones pulmonares en "halo" no se limitaban a los individuos histoplasmino-sensibles.

Acknowledgements

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The coccidioidin was supplied by Dr. Charles E. Smith of Stanford University, Palo Alto, California.

The histoplasmin was supplied by Dr. J. C. Peterson of Vanderbilt University, Nashville, Tennessee.

Bacteriologic studies were performed by Mr. E. A. Brosbe under the supervision of Dr. Wray J. Tomlinson, Chief of Laboratory Service, Veterans Administration Hospital, Fort Logan, Colorado.

The authors desire to express their sincere appreciation to these workers for their assistance in this study.

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TUBERCULOUS PERICARDITIS[1]

Report of a Case With a Ten Year Follow-up

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INTRODUCTION

Complete recovery from tuberculous pericarditis with effusion is sufficiently unusual to warrant the reporting of a single case.

Tuberculous pericarditis carries with it a mortality sufficiently high to question recovery in any given case. There appeared in the American medical literature in the ten years from 1937 to 1947 nine reports (1, 4, 6, 7, 8, 9, 10, 11 and 13) of individual cases of tuberculous pericarditis, and five series of cases (2, 3, 4, 5 and 14).

All of the 9 reported cases died. Hodges (4) found 5 cases in the records of the Norfolk General Hospital between the years 1923 and 1937, all of whom died. Of Keefer's (2) 20 cases, 18 died. Fenger and Hansen (14) collected 6 cases from the files of the Glen Lake Sanitarium, of whom 3 died. Four of Blalock and Levy's (5) 20 cases were alive at the time of writing (1937) but one of those was dying. Harvey and Whitehall (3) collected 95 cases from the records of the Johns Hopkins Hospital dating back to 1899. Of this series, 20 were cases of tuberculous pericarditis with effusion, of whom 16 died.

The aphorism that tuberculosis of the pleura and peritoneum is most often seen under forty years of age while tuberculosis of the pericardium is most often seen in those over forty years (2) of age must be modified in the light of recent findings.

Bellet *et al.* (12) found that tuberculous pericarditis occurred most often in children and young adults while half of Fenger and Hansen's (14) cases occurred between the ages of 20 to 40. Harvey (2) states that the disease is found in every decade of life.

It is generally agreed that the great preponderance of cases occurs in the Negro race. Of the 9 collected cases, 6 (1, 4, 6, 7, 11 and 14) were Negroes. In Harvey and Whitehall's (2) series of 20 cases, 15 were Negro.

Primary tuberculosis of the pericardium is extremely rare if not highly improbable (10). Tuberculous pericarditis is always secondary to some other tuberculous lesion (3). Tuberculosis of the pericardium arises from an invasion of tubercle bacilli from mediastinal lymph, the lungs, the pleura, or the peritoneum.

The condition is not always recognized except where massive effusion exists. In a series of 1,292 autopsies, Fenger and Hansen (14) found 22 cases of proved tuberculous pericarditis; 16 cases diagnosed at autopsy were not clinically diag-

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nosed. They conclude that tuberculous pericarditis is a rare complication of tuberculosis and carries a grave prognosis.

Keefer (2) subdivided his cases into the following groups:

(*1*) patients with symptoms and signs of a wasting disease and obscure fever;
(*2*) patients with symptoms and signs suggesting congestive heart failure;
(*3*) patients with symptoms and signs of multiple serous membrane tuberculosis;
(*4*) patients with symptoms and signs of massive pericardial effusion; and
(*5*) patients with symptoms and signs of miliary tuberculosis.

The course of the disease runs in three stages (10): sero fibrinous exudate; pericardial thickening and effusion; and the development of extensive adhesions confining the expansion of the heart. The average duration of the disease from the onset of symptoms until termination was three months (two to four months in Keefer's series (2) and three and seven-tenths months in Harvey and Whitehall's (3) series).

The poor prognosis in the older patients is associated with the inability of the cardiovascular system to cope with its added burden, while in the younger cases the frequency of development of other serious tuberculous lesions is responsible (3).

Especially in young people, tuberculous pericarditis with effusion must be differentiated from rheumatic pericarditis with effusion. Bellet *et al.* (12) noted the importance of finding the tubercle bacillus in the pericardial fluid, the absence of cardiac murmurs, and the demonstration of a small heart after the production of a pneumopericardium.

The following case is reported as it represents an apparently complete recovery from tuberculous pericarditis with effusion.

CASE REPORT

W. M., an 18 year old white male, was admitted to the Swedish Hospital (service of the late Dr. James Pullman) on August 4, 1936 complaining of pain in the chest and left shoulder, fever, and sweating.

In March, 1936 the patient had had a right pleural effusion. He was in bed for three weeks during which time his chest was tapped by one of us (J. A. C.) and about 2,000 cc. of fluid removed.

On July 29 the patient complained of suprasternal pain. He remained at home for the next two days, but on August 1 he went to the beach. The next day he complained of pain in the left chest which radiated to the axilla and the left shoulder. On August 4, because of an increase in the severity of the pain, fever, and rapid pulse he was hospitalized. Physical examination on admission to the hospital revealed an acutely ill young white male with a temperature of 104°F., a pulse rate of 130 and respirations of 40 per minute. On percussion the left anterior chest was dull from the third rib to the base. In the axilla the dullness merged with cardiac dullness. The breath sounds were diminished. The right chest was dull on percussion from the sixth rib to the base with diminished breath sounds. Posteriorly the left chest was dull at the base with diminished breath sounds. The right chest was flat posteriorly with absent breath sounds.

The left border of the heart could not be outlined by percussion as it merged with axillary dullness. The cardiohepatic angle was not diminished. Over the precordium the heart sounds were diminished and were best heard over the fourth costochondral junction. A systolic murmur was present.

Roentgenographic examination on the day of admission revealed that the trachea was in the midline and the heart enlarged in both borders. A pericardial effusion was present. The left diaphragm and the left lung were normal. The right diaphragm was not well outlined. There was an effusion of the right lower pleural cavity but the right lung was normal.

On admission the erythrocytes were 4,500,000 per cu. mm., the hemoglobin was 75 per cent of normal, the total leukocyte count was 11,700 per cu. mm. with 87 per cent neutrophils. Examination of the urine revealed no abnormalities.

During the next few days the patient's condition remained unchanged. Morphine was given for pain and restlessness. He became cyanotic though dyspnea was moderate. On August 5, a friction rub was heard over the fourth left costochrondral junction. The following day cyanosis was marked and there was some precordial pain. His pulse was irregular and of small volume. The friction rub was now heard over the entire precordium. There was slight venous distention, especially of the jugular veins. The dullness in the left axilla had diminished while the findings in the right chest were unchanged. A pericardial paracentesis was done and 225 cc. of serosanguineous fluid were removed.

During the next five days the patient remained seriously ill with fever (104°F.), auricular fibrillation, venous distension, cyanosis, and dyspnea. Six hundred cc. of straw-colored fluid were removed from the pericardial cavity on August eleventh. Examination of smear of the fluid was negative for acid-fast bacilli. A guinea pig was inoculated with the fluid. On October 10 the guinea pig was opened and examined by Dr. Leo Meyer. He reported

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FIG. 1. Chest roentgenogram taken August 3, 1936. The cardiac shadow is markedly enlarged in all directions. It is rounded, it has lost its normal border contours and has a configuration characteristic of pericardial effusion. There also is an effusion in the right pleural space, fluid reaching up to the level of the fourth rib and axilla, separating the parietal and visceral pleura and extending into the interlobar fissure between the right lower and middle lobes. The outer third of the right diaphragm is elevated and fixed.

FIG. 2. Roentgenogram obtained on October 7, 1944 shows that the cardiac contour has decreased greatly in size. The protrusion of the right cardiac border is gone. The right cardiac contour is normal. The left cardiac border still shows some elongation, as by a fixed pericardiopleural or mediastinopleural adhesive pleurisy, but the total size of the heart shadow has returned to approximately normal. The right diaphragm remains elevated and fixed in its outer margin. The right parietovisceral pleura are separated and thickened. There also are some fibro-proliferative changes now evident in the left lower lung field.

FIG. 3. Roentgenographic examination of the heart and lungs (December 1, 1947) in the postero-anterior, lateral and right and left oblique projections shows the following findings:

The cardiac shadow shows elongation of its left border in the oblique projections. There is an obliterative adhesive pleurisy involving the right diaphragm and the right axillary area. The parietovisceral pleural space in each axilla has been obliterated. The right diaphragm is elevated and fixed in its outer third. The left diaphragm is also partially fixed. There are dense basilar mediastinal pleural adhesions and there is a thickening and irregular calcification of the pleura postero-laterally in the lower third of the left chest. The parietovisceral space in the left chest has been obliterated by thickening of the pleura.

In view of the history of a previous tuberculous pleurisy, mediastinitis and pericarditis, these diffuse thickenings of the pleura probably represent the obliterative end stage of a tuberculous pleurisy. The calcific deposit within the pleura further bears this out. Judging by the present findings, the process has involved the pleural areas in each hemi-thorax, the mediastinal pleura and the pericardial pleura.

There is no evidence of parenchymal tuberculous deposit other than an occasional small, well-calcified primary focus.

"The inguinal glands were enlarged and caseous. Direct smear of the necrotic tissue discloses the presence of numerous acid-fast bacilli."

During the next two weeks the patient remained critically ill with considerable evidences of cardiac insufficiency. The pericardial fluid reaccumulated so rapidly during this period that it was necessary to remove 400 to 1,700 cc. on four separate occasions.

Gradual improvement was apparent, however, and by August 28 the patient was quite comfortable, although still seriously ill. There was no cyanosis or venous engorgement. His respirations were 30 per minute but his pulse still ranged about 130 per minute. Anteriorly there was flatness over the third rib merging with cardiac dullness. Examination of the chest on September 6 was as follows: The flatness over the left rib merged with cardiac dullness; the left axilla was resonant and breath sounds were heard; posteriorly there was impaired resonance from the sixth rib to the base, tubular breathing was present over this area; the heart rate was 110 per minute; there was evidence of pericardial fluid. By the 9th of September the area of dullness over the anterior left chest was smaller. The left axilla was completely resonant. There was no friction. Voice sounds were increased over the posterior chest. Two days later his pulse continued out of proportion to his temperature and was 130 per minute. However, his color was good and respirations were easy and normal. There was still evidence of pericardial fluid.

On September 21 the Mantoux test was negative. Improvement continued throughout the month and by October 30 his pulse was 86 per minute and it was possible to discontinue the digitalis.

The report of a chest roentgenogram obtained on October 31 read as follows: "The trachea is in midline; the base of the heart is enlarged; cardiac silhouette is greater than normal but not as great as on previous examination. I believe if any effusion is present in the pericardium the amount is small. There is some congestion at the right base with a very small amount of fluid in the left costophrenic area. Lungs show no definite indication of tuberculosis."

The patient was discharged on October 31, 1936 and remained at home in bed until February 15, 1938. During this fifteen and one-half month period the patient took his pulse daily and refused to leave his bed. Around February 15 he got out of bed for about an hour, took his pulse, was dissatisfied and returned to bed for three weeks. He then left his bed for short periods at a time, gradually increasing his stay out of bed. On October 15, 1938 he left the house for the first time. In November he played nine holes of golf and when he noted that his pulse quickened he returned to bed for a week. He returned to school at the end of 1938 and was graduated at the end of 1942. On six occasions he was called and examined by his draft board. At first his history was sufficient for rejection but, since the lad appeared so well and since physical examination revealed nothing, the hospital record was requested and a note signed by one of us (J. A. C.), certifying to the above illness, was sufficient to keep him from service. In 1946 he married.

From 1938 to date he has been seen by us for mild upper respiratory infections on about eight occasions. His chest has remained clear and there has been no sign of pericardial involvement or any other evidence of tuberculosis.

Acknowledgment

The writers wish to express their thanks to Dr. Alexander Sarafianos, who performed the pericardial taps.

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STREPTOMYCIN IN THE TREATMENT OF TUBERCULOUS PERICARDITIS[1]

Report of Three Cases

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INTRODUCTION

Tuberculous pericarditis is a very serious disease. In the 37 proved cases reported by Harvey and Whitehill (1) in 1937 from the Johns Hopkins Hospital there was an 83 per cent mortality, most of the cases dying within months of the onset of symptoms. Some (2) have stressed the occurrence of the disease in elderly males, and a few reports (3, 4) are to be found describing cases exclusively in males over 50 years of age. This, however, is not unique and might even happen in pulmonary tuberculosis. The Negro race is especially susceptible, particularly the age group 15 to 30 years which constitutes the majority of cases in some reported series (1, 5).

There is much discussion in the literature about "primary" tuberculous pericarditis (6). This term may be used in one of two senses. (*1*) "Clinically primary" tuberculous pericarditis (7, 8) means that the pericarditis is the patient's principal illness; it is the presenting disease and is the cause of his seeking medical aid. He may, in addition have demonstrable tuberculosis elsewhere. (*2*) The term "anatomically primary" tuberculous pericarditis means that tuberculosis is to be found only in the pericardium (3, 9). Some deny the existence of this form. The question is academic, however, for it would not be anticipated that the disease would originate in the pericardium. Tuberculosis may reach the pericardium by several routes. Most frequently the source of infection is from caseous mediastinal nodes lying in close proximity to the pericardium. Less frequently the disease is due to direct extension from tuberculosis of the pleura or lung, and still less common is hematogenous seeding of the pericardium.

The treatment of tuberculous pericarditis has been symptomatic, covering all that is implied in a sanatorium regimen. Pericardial tapping is done as necessary to relieve cardiac tamponade. To tap the pericardium and replace the fluid by air when tamponade is not present has been recommended by those who believe this may result in quicker recovery, or at least recovery without adhesions and without the possibility of a later chronic constrictive pericarditis. This assumption is still without proof. To the writers' knowledge there are only two reports in the literature on the use of streptomycin in tuberculous pericarditis. One report (10) concerned a 19 year old Negro girl who developed an acute pericardial effusion during the third month of a rapidly progressing tuberculosis of lymph nodes. Tubercle bacilli were cultured from the pericardial fluid. There was immediate improvement with the use of streptomycin (3 Gm. daily for 120 days)

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as shown by the complete disappearance of symptoms, and the cardiac silhouette decreased in size. Eleven months after streptomycin treatment the patient was still asymptomatic, had negative cultures, and was ambulatory at home. The cardiac silhouette was still slightly larger than normal. Meredith (11) reports the case of a Negro male aged 17 years who was febrile and had proved tuberculosis of the lymph nodes. A large pericardial effusion was tapped but tubercle bacilli were not recovered. After streptomycin was started (200 mg. every three hours for 33 days) there was marked improvement. The patient became afebrile, gained weight, and the cardiac silhouette decreased in size. Three months after streptomycin therapy the patient was feeling very well, although he was still in a sanatorium for treatment of his pulmonary tuberculosis.

The present report is concerned with 3 patients with tuberculous pericarditis who received streptomycin as part of their treatment.

The first case is that of a middle aged white male physician who had an acute tuberculous pericardial effusion followed in a few months by chronic constrictive pericarditis for which he was surgically treated. One year later he was in excellent health. The other two cases are of Negro youths. One (case 2) made a clinical recovery but there persists a markedly thickened, though asymptomatic, pericardium. The other patient (case 3) developed a pericardial effusion which later completely reabsorbed without apparent significant residual except for an abnormal electrocardiogram. The amounts of streptomycin used in the three cases were: 7.5 Gm. in 13 days; 120 Gm. in 120 days; and 42 Gm. in 42 days, respectively. In cases 1 and 2 tubercle bacilli were cultured from the pericardial fluid. In case 3 no aspiration was done and therefore the diagnosis of tuberculous pericarditis, though probable, was not proved. None of these patients showed any toxic symptoms from the streptomycin treatment.

CASE REPORTS

Case 1: A regular army physician, 59 years of age, suddenly became ill in October, 1945 with malaise, fever, and weakness following a mild upper respiratory infection. The diagnosis was left basal pneumonia. In two weeks he was afebrile and ambulatory but after three days his fever recurred. Penicillin and sulfonamides were now without benefit. In December, 1945 he was admitted to Letterman General Hospital where a diagnosis of pericardial effusion was made on the basis of increased cardiac dullness, faint heart sounds, distended jugular veins, and a liver enlarged downwards 3 cm. below the right costal margin. Paradoxical pulse was present, blood pressure was 120 systolic and 80 diastolic, venous pressure (antecubital) 177 mm. water. The total leukocyte count was 10,000 per cu. mm. with 74 per cent segmented cells. His pericardial sac was tapped and straw-colored fluid removed, which showed a few white blood cells but no organisms on smear, a specific gravity of 1.032, and 5.5 Gm. of protein per 100 cc. On January 25 this pericardial fluid was reported positive for tubercle bacilli on culture and guinea pig inoculation.

Beginning on February 2, he was given 0.1 Gm. streptomycin intramuscularly every four hours for a total of 7.5 Gm. This was followed by a decrease in the amount of pericardial fluid, a drop in the erythrocyte sedimentation rate from a high of 95 mm. in one hour to 12 mm. by March, 1946, and a drop in venous pressure from 177 mm. to 120 mm. of water, with improvement in his general health. Search for other manifestations of tuberculosis showed that the patient had been hospitalized in 1942 because of a chronic cough of four years duration. Mild hypertension and arteriosclerosis were found, and in

addition, a markedly thickened and calcified left pleura with some nodose fibrous lesions in the left apex. The diagnosis at that time was arrested pulmonary tuberculosis, and the patient was returned to limited duty.

On March 29, 1946 he was transferred to Moore General Hospital because of his pericardial and pulmonary tuberculosis. He very soon developed marked enlargement of the liver, ascites, peripheral edema, and rales in the bases of both lungs. A diagnosis of chronic constrictive pericarditis was made and he was sent to Fitzsimons General Hospital in May for surgical treatment. Here all the evidence necessary for this diagnosis was confirmed: distended neck veins showing inspiratory filling; distant heart sound; pulsus paradoxicus; ascites; a nontender liver palpable four fingerbreadths below the right costal margin; and a venous pressure (antecubital) of 180 mm. water. Electrocardiograms were available for the period prior to this illness, and they were normal. Tracing of December 4, 1945 showed a lowered amplitude of QRS and T in all leads taken: I, II, III and CF4. By December 31 there was asymmetrical inversion of T in leads I, II, III and a diphasic T in CF4.

The preliminary treatment consisted of a low salt diet, digitalis, and mercurial diuretics. He showed temporary improvement, but after August, 1946 he became worse and his ascites increased. In January, 1947 a pericardiectomy was done and 7 thick pieces of laminated fibrous tissue were removed with immediate improvement of cardiac function. Histological study of the excised material showed a relatively acellular dense fibrous tissue with discrete granulomatous nodules containing either epithelioid cells, lymphocytes and an occasional Langhans' giant cell, or relatively acellular encapsulated organizing caseous foci. No acid-fast bacilli were demonstrated.

Following the operation, the patient improved but it was one year before ascites and edema of the legs were fully controlled. His electrocardiogram remained abnormal. The patient was in fine condition July, 1948 having made an apparently complete recovery.

Case 2: A 20 year old Negro soldier was admitted to William Beaumont General Hospital in December, 1946 stating that he had been well until one week previously when he noted the onset of malaise, a nonproductive hacking cough, and a constant dull ache in the left scapular region which was not aggravated by deep breathing or coughing. He then developed a sharp pain over the right chest anteriorly that was aggravated by deep breathing and coughing. The patient had been working as a butcher and laborer in an army hospital for the previous eleven months. His past and family history were noncontributory and there was no known exposure to tuberculosis.

Physical examination on admission to the hospital revealed that he was well developed and well nourished, and he appeared acutely ill. His blood pressure was 120 systolic and 80 diastolic. Examination of the heart revealed that the apex beat was diffuse and the left border of dullness on percussion was on the anterior axillary line in the fifth intercostal space. A soft systolic murmur was heard in the left third interspace close to the sternum. The second pulmonic sound was reduplicated and the rhythm was normal. The remainder of the physical examination was negative. Laboratory data on admission were as follows: total leukocyte count 6,500 per cu. mm. with 45 per cent segmented neutrophils, 9 per cent band cells, 43 per cent lymphocytes, and 3 per cent eosinophils. The hemoglobin concentration was 15.6 Gm. per 100 cc. Urinalysis was normal and the specific gravity was 1.020. A serologic test for syphilis (Kahn) was negative.

For the first two weeks the patient presented mainly a febrile course, with temperatures ranging from a low of 100°F. to a high of 103°F., spiking daily in the afternoon. The pulse averaged 90 to 100 per minute. He complained mainly of headaches. On December 17, fluoroscopy revealed an enlarged cardiac shadow and an electrocardiogram showed slight elevation of the ST segments in leads I, II, III and CF4 (figure 3). On December 21, a pericardial friction rub was heard and an electrocardiogram showed inversion of T in leads I, II and CF4. By December 26, the pericardial fluid had increased and the heart tones were of poor intensity. The erythrocyte sedimentation rate was 38 mm. in one hour.

On February 3, 1947 a pericardial tap was done with the removal of 100 cc. of dark straw-

colored fluid. From this tubercle bacilli were cultured, but they were nonpathogenic for a guinea pig. Smear of the fluid sediment was negative. On February 17, the patient was drowsy, weak and confused. Cerebrospinal fluid examination was normal and neurological examination revealed a horizontal nystagmus with the fast component to the left. However, the patient was alert by February 22, and no definite cause for the episode was found, though it may have been salicylism. During this period albuminuria appeared and it persisted throughout his hospitalization. A second pericardial tap on March 1, yielded 250 cc. of dark amber fluid from which tubercle bacilli were again cultured which were still nonpathogenic for the guinea pig. The patient continued to have a remittent and intermittent fever up to 102°F. with a pulse rate of 90 to 100 per minute. His abdomen was slightly distended and "doughy" on palpation. On March 3, 1947 a chest roentgenogram revealed a small amount of fluid in both costophrenic angles. There was a gradual increase in the right pleural effusion and a thoracentesis was done on that side, with the removal of 330 cc. of straw colored fluid. No organisms were recovered on culture or guinea pig inoculation.

Sputum examinations for tubercle bacilli and blood cultures were negative. The sedimentation rate remained elevated at around 36 mm. in one hour. Pericardial taps on March 22, June 19 and July 21 and 28 yielded from 250 to 500 cc. of turbid yellow fluid (figure 1). Cultures of these specimens revealed tubercle bacilli but all reports obtained at the time were nonpathogenic for a guinea pig. At the end of July, the total plasma proteins were 7.9 Gm. per 100 cc. (albumin 2.8, globulin 5.1). There was no peripheral edema, the liver was not palpable, but the venous pressure (antecubital) was 230 mm. of water. The patient was given several injections of concentrated serum albumin. His temperature gradually fell over a period of several months and by September, 1947 he was having only occasional elevations to 100°F.

Because of the diagnosis of tuberculous pericarditis, the patient was transferred to Fitzsimons General Hospital on October 8, 1947 for further study. Examination at that time revealed signs of fluid in both lung bases, and a friction rub was heard over the entire precordium. On October 14, the patient was dyspneic and cyanotic and he complained of pain in the left anterior chest. The liver was palpable for the first time, four fingerbreadths below the right costal margin, and was tender. By October 17, there was pitting edema of both legs up to the knees and definite evidence of ascites. Venous pressure (antecubital) was 150 mm. of water, decholin time was 19 seconds, and ether time, 7 seconds. The patient was treated with mercupurin with loss of the edema.

On October 27, he was started on streptomycin, 0.5 Gm. intramuscularly every twelve hours, to be continued for 120 days. From this time on, the patient remained afebrile and his pulse ranged from 80 to 90 per minute. Erythrocyte sedimentation rates remained about 25 mm. in one hour. Total plasma proteins were 8.5 Gm. per 100 cc. (albumin 2.7 globulin 5.8). The patient had no edema or ascites by the middle of November. At this time a report was received on pericardial fluid removed on August 20 at William Beaumont General Hospital stating that it was positive for tubercle bacilli which were pathogenic for a guinea pig. Numerous gastric washings and urine cultures were negative for tubercle bacilli. White blood cell counts remained normal. On December 5, venous pressure (antecubital) was 135 mm. water with a rise to 200 mm. on pressure over the right upper quadrant of the abdomen. Arm-to-tongue circulation time (Decholin) was 22 seconds and with ether was 12 seconds. The patient was asymptomatic and had a good appetite by the end of December. During January, 1948 he continued this same good course and the only physical findings were a false pulsus paradoxicus (present only when he held a deep inspiration), enlargement of the cardiac area, and evidence of pleural thickening over the right lower lobe. The liver could not be felt.

During the rest of the patient's hospital stay until he was transferred to a Veterans Hospital in April, 1948 he remained asymptomatic and feeling well (figure 2). It was difficult to keep him on modified bed rest. The total plasma protein was 8.6 Gm. per 100 cc. (albumin 4.4, globulin 4.2). Retrograde pyelograms and roentgenograms of bone, as well as liver

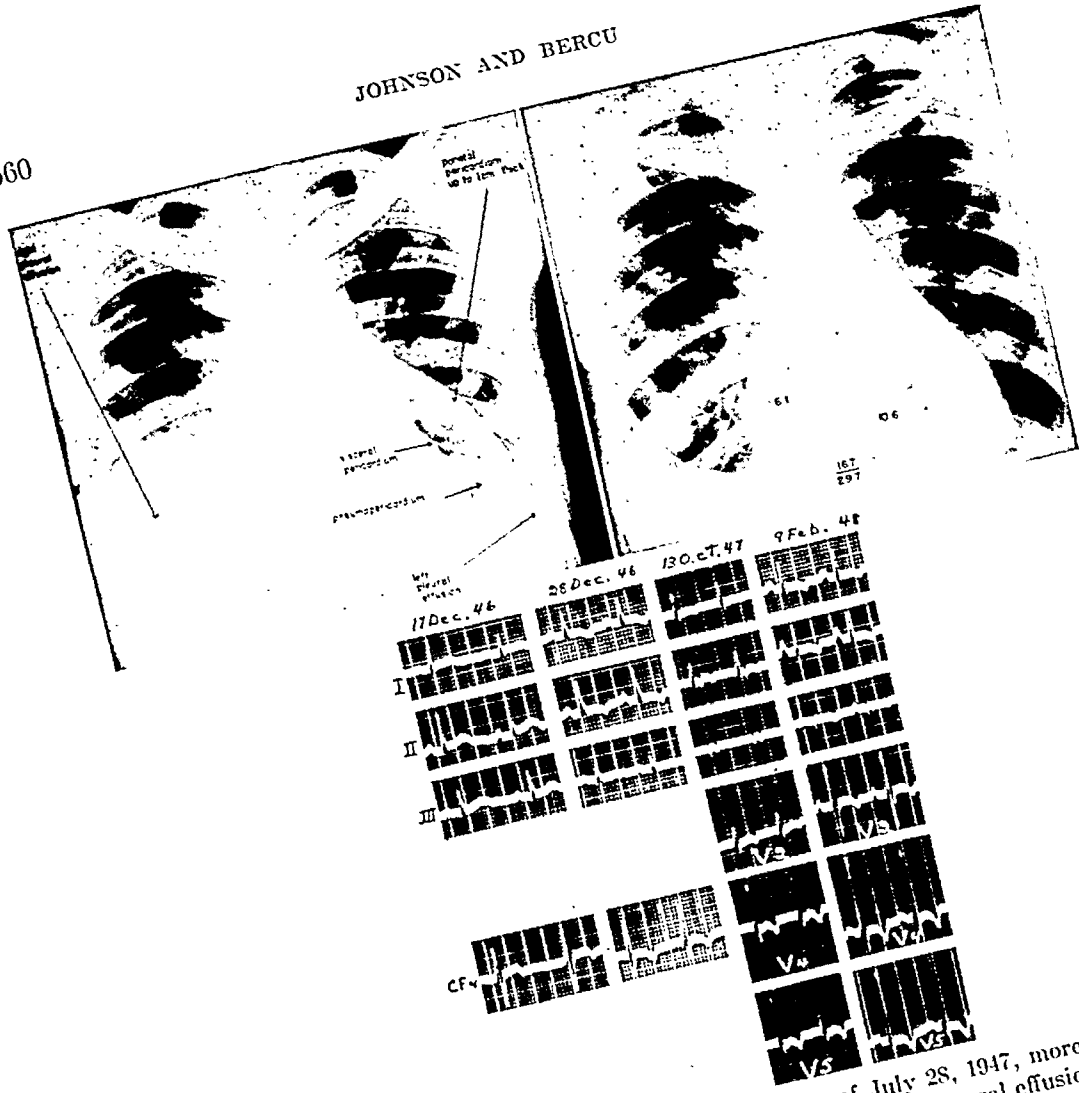


FIG. 1. Case 2. (Upper left) Chest roentgenogram of July 28, 1947, more than seven months after onset of illness, showing pericardial and bilateral pleural effusions. The air in the pericardium was injected after removal of fluid and demonstrates a greatly thickened parietal pericardium.

FIG. 2. Case 2. (Upper right) Chest roentgenogram of March 18, 1948 after the patient had been given four months of streptomycin therapy. There remains thickened pericardium and pleurae.

FIG. 3. Case 2. (Bottom) Electrocardiogram of December 17, 1946 showing slight elevation of ST segments in leads I, II, III and CF4. This is a pattern seen early in pericarditis and seldom obtained in tuberculous pericarditis. The three following tracings show the more commonly found pattern with inversion of T waves and decreased amplitude of QRS. Nodal rhythm is present in tracing of October 13, 1947; this was transient.

and kidney function tests, were normal. The false pulsus paradoxicus disappeared. Venous pressure (antecubital) was 140 mm. of water, and arm-to-tongue (Decholin) circulation time 17 seconds.[2]

[2] In January, 1949 this patient was still clinically well.

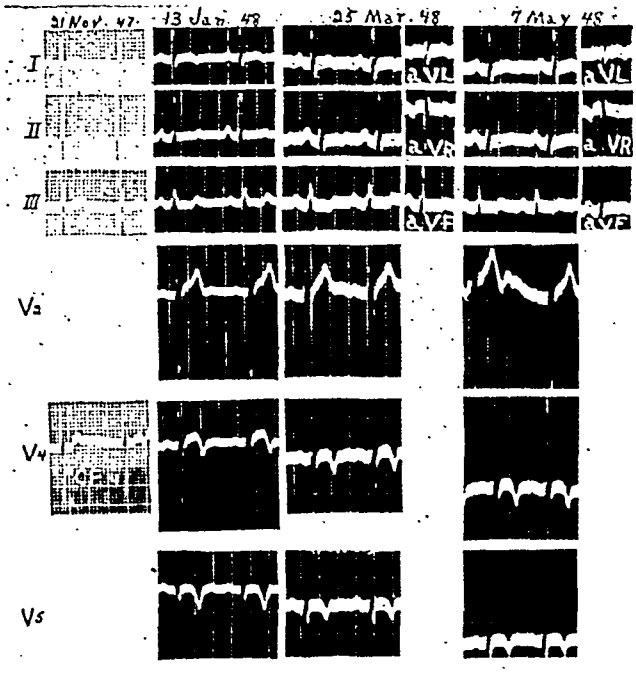
Case 3: An 18 year old Negro soldier was admitted to a station hospital in Japan on August 26, 1947, complaining of loss of weight for three months and chills for four days. The patient stated that during the past three months he had had poor appetite, lost 15 pounds in weight and felt tired. Four days before admission he began to notice chilliness at night necessitating the use of extra covering. He had a "tingling" sensation over the sternum. There was no cough or hemoptysis.

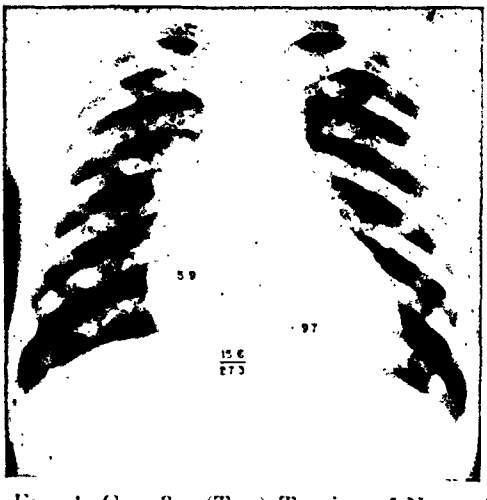
Past history was significant in that the patient had had a chancroid four months before admission which had been treated with sulfadiazine. Family history was noncontributory, and there was no history of exposure to tuberculosis.

Physical examination revealed him to be thin, poorly nourished, undersized, and he appeared chronically ill. Blood pressure was 110 systolic and 60 diastolic. The heart was not enlarged, the tones were of good quality, and no murmurs were audible. Pulse was 52 per minute with regular rhythm. The rest of the examination was negative. Laboratory data: erythrocyte count was 4.16 million per cu. mm., the hemoglobin was 100 per cent of normal, the total leukocyte count was 8,700 per cu. mm. with 60 per cent segmented neutrophils, 34 per cent lymphocytes, 5 per cent monocytes, and 1 per cent eosinophils. Erythrocyte sedimentation rate was 56 mm. in one hour. Serologic test for syphilis (Kahn) was positive. The urine was normal. Hookworm larvae and Trichuris trichuria ova were found in the stools. Examination of the blood for malarial parasites was negative. Blood cultures were sterile. A roentgenogram of the chest showed an enlarged right upper hilar lymph node and lines of increased density radiating into the first and second right anterior interspaces.

The patient had an intermittent fever with daily peaks of 103°F. He had only a feeling of malaise and complained of no pain. During the last half of September, the patient's fever became lower with daily rises to 99°F. He felt stronger and his appetite improved. The erythrocyte sedimentation rate remained about 35 mm. in one hour. The patient was treated for the hookworm and whipworm infestations with hexylresorcinol and tetrachlorethylene. On October 13, the titer of the serologic test for syphilis (Kahn) rose to 320 "units" and the patient was given 8 million units of penicillin intramuscularly over a period of ten days. Except for an initial Herxheimer reaction, the patient's fever remained low grade with rises to 99.5°F. On November 18, while the patient was sitting up, he was suddenly seized with a sharp pain in the left anterior chest which radiated to the left shoulder. There was marked dyspnea but no cough or hemoptysis. The pain disappeared spontaneously in about thirty minutes. The total leukocyte count was 5,800 cells per cu. mm. with a differential showing 9 per cent eosinophils. Sedimentation rate was 32 mm. in one hour. An electrocardiogram showed only sinus arrhythmia (figure 4). On November 19, a friction rub could be heard over the entire precordium. A soft blowing systolic murmur was heard at the apex. Chest roentgenogram revealed an increase in the size of the heart of 3 cm. in the transverse diameter. A tentative diagnosis of tuberculous pericarditis was made. The patient's temperature was now intermittent with daily spikes to 101°F. The pericardial friction rub disappeared by November 21 and the heart sounds became distant. The titer of the Kahn test dropped gradually to 40 "units". Sputum examinations for tubercle bacilli remained negative. Chest roentgenograms remained unchanged. On November 28, the patient was evacuated to the zone of the interior. On arrival in the United States and while awaiting transfer to an Army general hospital, laboratory study showed: a total leukocyte count 5,000 cells per cu. mm., hemoglobin concentration 14.8 Gm. per 100 cc., erythrocyte sedimentation rate 6 mm. in one hour, Kahn test 10 "units" and urine normal (figure 5).

On admission to Fitzsimons General Hospital January 8, 1948 the patient was asymptomatic. His temperature was normal. Physical examination revealed no abnormalities. The erythrocyte sedimentation rate was 10 mm. in one hour. A roentgenogram of the chest showed the large discrete lymph node enlargement in the right hilar region. The heart shadow was smaller but still enlarged. Lung fields were normal. Electrocardiogram showed





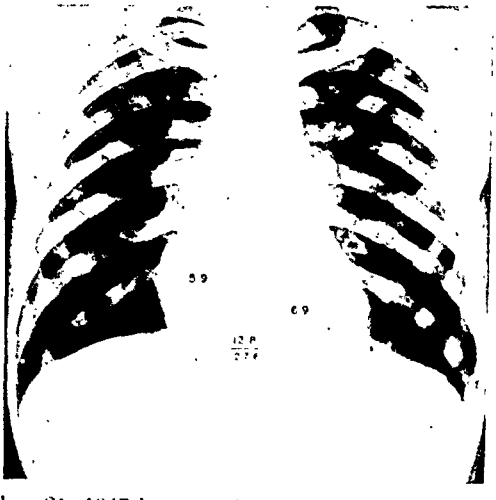


FIG. 4. Case 3. (Top) Tracing of November 21, 1947 is essentially normal though the notching of T in CF4 may be suspicious of pericarditis. Tracing of January 13, 1948 reveals a pericarditis pattern showing inversion of T in leads I, V_4 and V_5, and decreased amplitude of T in leads II and III. In March, 1948 further changes are seen in that T in leads II and III is now inverted. Tracing of May 7, 1948 shows regressive changes in T waves of limb leads. However, the abnormal T in V_4 and V_5 persists.

FIG. 5. Case 3. (Lower left) Chest roentgenogram of December 26, 1947 showing a pericardial effusion. Note especially the enlarged (right) hilar lymph node, a common precursor of tuberculous pericarditis.

FIG. 6. Case 3. (Lower right) Chest roentgenogram of January 26, 1948 showing return of the cardiac silhouette to normal size following streptomycin therapy.

inverted T in leads I, V_4 and V_5 and was interpreted as showing a pericarditis pattern (figure 4).

Because the lymph node enlargement was considered to be tuberculous and potentially unstable, and because of the associated pericarditis, the patient was given a course of streptomycin intramuscularly, 0.5 Gm. every twelve hours for a total of 42 days. During this time the patient remained asymptomatic. He was afebrile and his total and differential leukocyte count and erythrocyte sedimentation rate remained normal. Examinations of numerous gastric washings were negative for tubercle bacilli. The serologic tests for syphilis became negative. Electrocardiograms showed some regressive changes but inversion of T persisted in leads $V_{4,5,6}$ (figure 4). A roentgenogram of the chest on January 26, 1948 showed a moderate decrease in the size of the enlarged right hilar node. The cardiac silhouette had decreased to normal size (figure 6). In July, 1948 the patient was transferred to a Veterans hospital and, at that time, he was asymptomatic, afebrile, and appeared well. Physical examination revealed no abnormalities. The only apparent abnormalities remaining were the large hilar node and the electrocardiographic evidence of pericarditis.

SUMMARY

Two cases of proved and one case of highly probable tuberculous pericarditis which responded satisfactorily to streptomycin therapy are reported.

SUMARIO

La Estreptomicina en el Tratamiento de la Pericarditis Tuberculosa

Los tres casos comunicados, dos comprobados y uno muy probable, de pericarditis tuberculosa respondieron satisfactoriamente a la estreptomicinoterapia.

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STREPTOMYCIN IN EXPERIMENTAL TUBERCULOSIS[1,2]

III. Effect on the Pathogenesis of Early Tuberculosis in the Guinea Pig Infected with Streptomycin-sensitive H37 Rv Tubercle Bacilli

W. STEENKEN, JR. AND PHILIP C. PRATT

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INTRODUCTION

Up to the present time, reports of the results of streptomycin therapy in the experimental animal have dealt chiefly with therapy instituted immediately after injection with virulent tubercle bacilli, or therapy instituted after the disease was well established. This paper reports the early course of a tuberculous infection in normal guinea pigs to which streptomycin was administered prior to injection with highly virulent streptomycin-sensitive microorganisms.

It was demonstrated by Krause (1, 2), and later by others (3, 4, 5, 7), that within four to six days after subcutaneous inoculation there is a widespread dissemination of tubercle bacilli throughout the body of the guinea pig, although gross and microscopic pathological lesions may not be observed before six to ten days. Krause's findings show that, in order to prove the presence or absence of viable tubercle bacilli in early tuberculosis in the guinea pig, there should be a subinoculation of the triturated organs from an infected animal into normal guinea pigs, in addition to gross and microscopic examination of the tissues.

METHODS

In the experiment reported here, 32 tuberculin-negative male guinea pigs, each weighing about 450 Gm., were divided into two groups of 16 each, one group for treatment and one for control.

Preparation of suspension of bacilli: An evenly dispersed 14 day culture of H37 Rv microorganisms in Tween-albumin medium was diluted with physiological saline until a standard loopful of the culture spread on a slide over an area 15 by 15 mm. in size, contained 20 to 50 microorganisms per oil immersion field. The final suspension adjusted in the Klett Summerson colorimeter gave a dial reading of 17. Using this figure, it was calculated (6) that 1.0 cc. contained approximately 20,000,000 tubercle bacilli.

Therapy: The animals in both groups were injected subcutaneously in the midline, anterior to the symphysis pubis with 0.2 cc. of the suspension, which is equivalent to approximately 4,000,000 bacilli. In Group I, streptomycin therapy was begun forty-eight hours prior to infection. Each animal received 18,000 γ of the drug per day in four doses; three doses of 4,000 γ each at 7 a.m., at 12 noon, and 5:30 p.m., and the last dose of 6,000 γ at 10:30 p.m. The treatment was continued throughout the experiment.

Sacrifice periods of animals and subinoculation: Two treated and two control animals were killed at 3, 5, 10, 15, 21, 27, and 30 day intervals. Tissues from the treated animals sacrificed at 3, 5, 10, 15, and 21 days, as well as tissues from the control animals killed at 3, 5, and 10 days, were removed and studied as follows: One iliac node, half of the spleen, and two

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[1] From The Trudeau Laboratory of The Trudeau Foundation for the Clinical and Experimental Study of Pulmonary Disease, Trudeau, New York.

[2] This study was aided by a grant from the Division of Research Grants and Fellowships of the National Institute of Health, U. S. Public Health Service.

tracheobronchial nodes were removed aseptically from each animal. These were ground separately in a mortar with the aid of a pestle and the material suspended in a minimum of physiological saline. Each triturated organ was injected subcutaneously into the inguinal region of a normal guinea pig. These subinoculated animals were then followed by monthly tuberculin tests and autopsied as their reactions became positive, or after a four month period, whether they were positive or negative.

RESULTS

Tuberculosis by subinoculation was produced by each of the iliac nodes and by the spleens obtained from both the infected controls and the treated animals killed at the end of 3 days. All tissues obtained subsequently from the same organs of the animals killed at various intervals likewise produced tuberculosis when inoculated into normal guinea pigs.

The triturated bronchial nodes obtained from the treated pigs and the controls at the end of 3 and 5 days, when injected into normal guinea pigs, failed to produce tuberculosis. The bronchial nodes obtained from two infected controls, when subinoculated at the end of 10 days, produced progressive tuberculosis in normal guinea pigs, whereas the nodes obtained from the treated animals failed to do so. The tracheobronchial nodes from the two treated animals killed at 15 days, however, did produce tuberculosis on subinoculation. At 21 days, the tracheobronchial nodes from one of the treated animals produced tuberculosis on subinoculation, but those from the other failed to do so.

Pathologic Changes

Recordings of gross observations on all animals were made at autopsy. The spleen, liver, lungs, and the tracheobronchial and iliac nodes from each animal were then fixed in Zenker's solution and stained with hemotoxylin and eosin for microscopic study. The essential pathologic findings are presented in table 1, in which the gross and microscopic observations of the various organs are recorded according to the duration of the disease. As previously stated, two treated and two control animals were killed at each sacrifice period. The lesions in each of the animals within each group were, in most instances, comparable. When the described lesion appeared in only one of the animals, this fact is indicated by an asterisk.

In relation to the controls, it is evident that the treated animals exhibited little disease, that fewer organs were involved, and that, with one exception, the lesions were less extensive. The exception occurred in the iliac nodes of the animals sacrificed at 10 days; the extent of the disease was identical in each animal of the two groups and the lesions covered several microscopic fields.

It is important to emphasize the fact that lesions were observed in the spleen and lungs, although these lesions were only microscopic. The livers also might have revealed lesions had serial sections been made on this organ. A portal scar was observed in one of the treated animals killed at 27 days; this probably represented a healed tuberculous lesion. The iliac nodes of the animals killed at 27 days showed caseation.

The foci of lymphocytes seen in the lungs of the treated animals sacrificed

TABLE 1

Gross and Microscopic Pathological Findings in Streptomycin-treated and Nontreated Animals in Relation to Time after Infection

Sacrifice periods	Type of pathology	Treated animals: Iliac node	Spleen	Lung	Liver	Tracheobronchial node	Control animals: Iliac node	Spleen	Lung	Liver	Tracheobronchial node
7 days	Gross	Slight enlargement	0	0	0	0	Slight enlargement	0	0	0	0
	Microscopic	0	0	0	—	0	0	0	0	—	0
5 days	Gross	Slight enlargement	0	0	0	0	Slight enlargement	0	0	0	0
	Microscopic	0	0	Foci of lymphocytes*	—	0	0	0	0	—	0
10 days	Gross	Enlarged	Slight enlargement	0	0	Slight enlargement	Enlarged	Enlarged with tubercles	0	0	Enlarged
	Microscopic	Macrophages in sinuses	0	Foci of lymphocytes	—	0	Macrophages in sinuses	Macrophages	Macrophages	—	0
15 days	Gross	Enlarged	0	0	0	0	Tubercles	Tubercles	Tubercles	Tubercles	Tubercles
	Microscopic	Macrophages in sinuses	0	0	—	0	Macrophages, epithelioid tubercles and necrosis	Macrophages and epithelioid tubercles	Epithelioid tubercles	Epithelioid tubercles	Epithelioid tubercles
21 days	Gross	Enlarged	0	Translucent foci**	0	Slight enlargement	Tubercles	Tubercles	Tubercles	Tubercles	Tubercles
	Microscopic	Macrophages and necrosis	Macrophages	Foci of lymphocytes	0	0	Macrophages, epithelioid tubercles and caseation	Macrophages, epithelioid tubercles and caseation	Macrophages, tubercles and caseation	Epithelioid tubercles with caseation	Macrophages, epithelioid tubercles

27 days	Gross	Tubercles	0	0	0	0	Caseous foci	Tubercles	Tubercles	Tubercles	Tubercles
	Microscopic	Epithelioid tubercles with caseation	0	Epithelioid tubercles and foci of macrophages	Small scar**	0	Epithelioid and caseous tubercles	Necrosis, epithelioid tubercles and caseation	Macrophages, epithelioid cells and caseation	Epithelioid tubercles and caseation	Epithelioid tubercles and caseation
39 days	Gross	Slight enlargement	0	0	0	0	Caseous tubercles	Necrotic	Tubercles	Tubercles	Caseous tubercles
	Microscopic	Few macrophages and scar	0	0	0	0	Epithelioid and caseous tubercles	Macrophages, epithelioid tubercles and necrosis	Macrophages and caseation	Epithelioid and caseous tubercles	Epithelioid and caseous tubercles

* See text. ** Noted in only one of the animals.

PLATE I.

Magnification of cross sections of spleens ×10

FIG. 1. Treated animal 15 days after infection.
FIG. 2. Treated animal 21 days after infection.
FIG. 3. Treated animal 30 days after infection.
FIG. 4. Control animal 15 days after infection.
FIG. 5. Control animal 21 days after infection.
FIG. 6. Control animal 30 days after infection.

Note the increased size of the spleens of the control animals with progressing destruction of the malpighian bodies at 15 and 27 days, and the areas of necrosis at 30 days. Compare the relatively constant and normal size of the spleens of the treated animals. Microscopic evidence of tuberculosis was present in the spleen of the treated animals at 21 days (See Plate III, figure 2).

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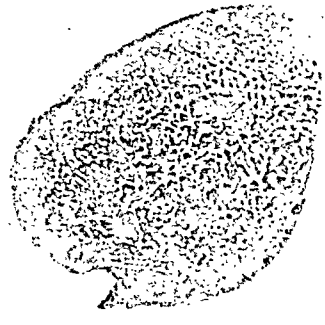

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PLATE II.

Magnification of sections of iliac lymph nodes

FIG. 1. Treated animal 21 days after infection. ×10.

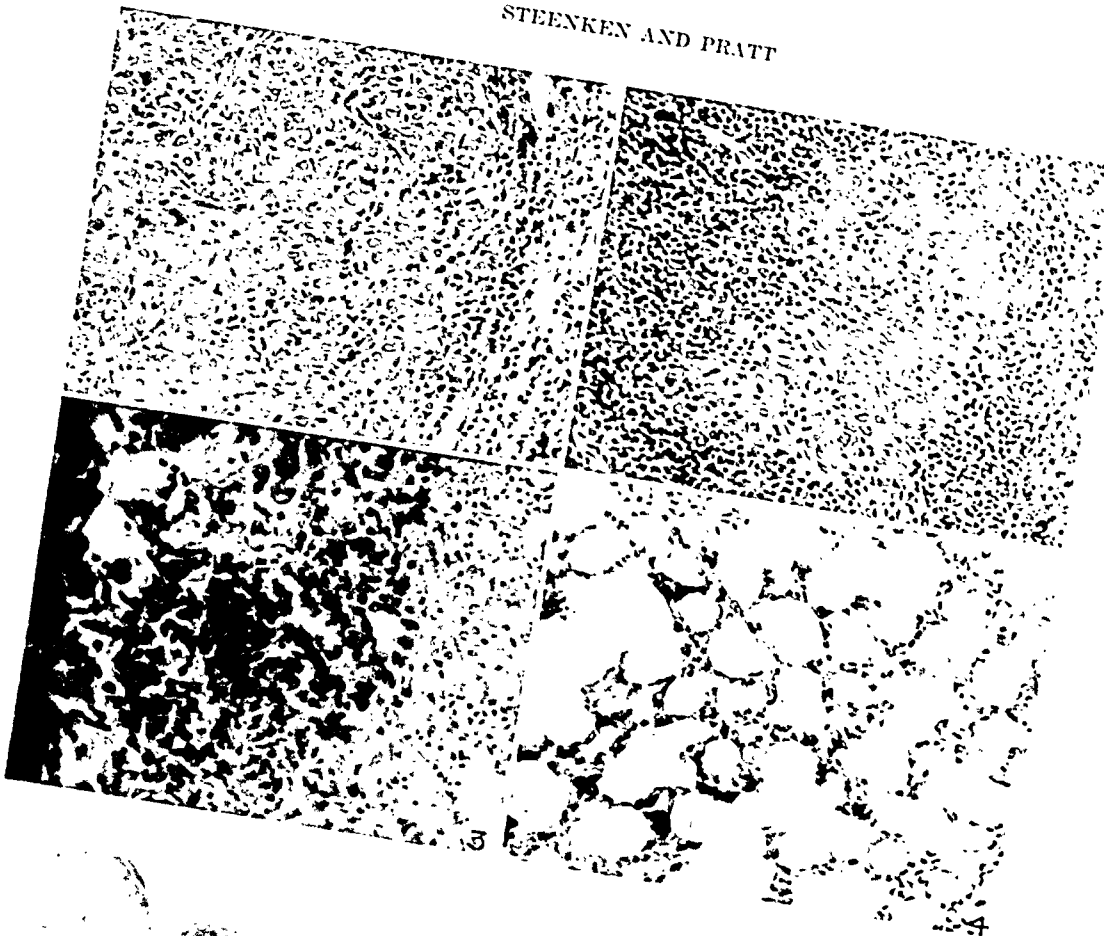
FIG. 2. Control animal 21 days after infection. ×10.

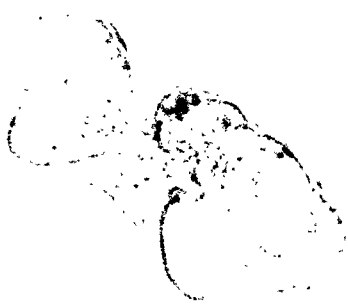
FIG. 3. Control animal 39 days after infection. ×10.

FIG. 4. Treated animal 39 days after infection. ×10.

Note the larger size of the iliac node in the control animals, also the lesions, with caseation, in the node from the animal treated 21 days, and the absence of a lesion at 39 days.

FIG. 5. Higher magnification of node shown in figure 1. Note infiltration with macophages and suggestion of formation of epithelioid cells. ×160.





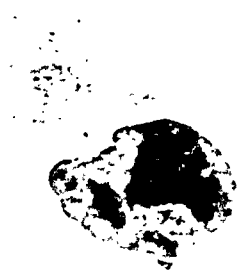


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PLATE III

FIG. 1 Section of spleen from control animal at 21 days ×160 Note destruction of malpighian body by macrophages and epithelioid cells. The margin of the malpighian body is at the right

FIG. 2 Section of spleen from treated animal at 21 days. ×160 Note the infiltration within a malpighian body of macrophages, and epithelioid cells

FIG. 3 Section of lung from control animal at 30 days ×160 Note tuberculous pneumonia

FIG. 4 Section of lung from treated animal at 30 days ×160 Note absence of lesion

FIG. 5 Section of tracheobronchial node from control animal at 30 days ×10 Note enlargement and loss of structure due to the presence of epithelioid tubercles and caseation

FIG. 6 Section of tracheobronchial node from treated animal at 30 days ×10 Note absence of lesion. Node is of normal size.

on the fifth and tenth days were very conspicuous; they were compact, and many of them had germinal centers, evidence which indicated that they could be considered true lymphoid follicles. While such lymphoid foci are not normally present in the lung of the guinea pig, they were found rather frequently. The reason for their presence is not apparent. Little significance is attached to them, since in other experiments there has been no regular occurrence of such lymphoid foci in the lungs of treated animals. Aside from these lymphoid foci, the microscopic appearance of the lesions in the two groups was similar, and apparently streptomycin did not affect the cellular reaction to the bacilli (See figures).

DISCUSSION

The results of this experiment reveal that streptomycin therapy begun forty-eight hours prior to injection subcutaneously with virulent organisms did not prevent the distribution of tubercle bacilli in the body of the guinea pig. The fact that tubercle bacilli were demonstrated in the tracheobronchial lymph nodes of the control animals by subinoculation at 10 days, but not until 15 days in the treated animals, is not considered significant because this variation is within the range reported by Krause (1, 2). However, this finding may be contrasted with Krause's observation that in the immunized guinea pig the dissemination of bacilli from the site of inoculation is delayed. These observations indicate that the effect of treatment with streptomycin does not duplicate this phenomenon of host-acquired resistance inasmuch as streptomycin limits the multiplication of tubercle bacilli but does not result in the localization of the organisms at the site of inoculation.

The difference in the amount of tuberculosis observed in the two groups is striking and needs no further comment, for the dramatic effect of streptomycin on experimental tuberculosis in the guinea pig is well known. Nevertheless, the fact that tuberculous lesions developed in the treated animals deserves the closest study.

A review of the course of the infection in the treated animals shows that lesions were first seen in the iliac nodes at 10 days. Progression of this lesion was slow but definite up to 27 days, at which time caseation was present. At 39 days, the next sacrifice period, only a few macrophages and a small area of scar tissue were present. In the spleen, foci of macrophages were observed in the malpighian bodies at 21 days, but not before or after this period. At 27 days, small groups of macrophages and a few epithelioid tubercles were observed in several of the lymphoid foci in the lungs. At 39 days, both these and the lymphoid foci were absent, and the lungs appeared to be normal. It is apparent then that, despite streptomycin therapy, the tuberculous lesions progressed for a period of about 27 days, then regressed, and at 39 days the only lesions observed were a few macrophages and a scar in the iliac node.

The presence of caseation indicates that the animals had become hypersensitive. The animals killed at 27 and 39 days were skin tested with 5 per cent Old Tuberculin and both the control and the treated animals showed strong

positive reactions. Since it is known that hypersensitivity and acquired resistance appear at about the same time (9, 10), it is assumed that these animals had developed some resistance at this time. Thus, the cessation of progression and the appearance of a regressive tendency of the lesions in the treated animals coincided with the development of acquired resistance.

The fact that the treated animals became hypersensitive to tuberculin is also significant. It has been established that very small numbers of dead tubercle bacilli will not produce skin hypersensitivity when injected into guinea pigs subcutaneously. Apparently, the organisms multiplied in the animals despite streptomycin therapy.

The above considerations lead us to believe that streptomycin acts in conjunction with acquired resistance in the guinea pig, and that acquired resistance plays an important part in the dramatic results seen when an established tuberculosis in the experimental animal is treated with streptomycin.

Further experiments are in progress to study the early course of tuberculosis in guinea pigs immunized with living attenuated H37 Ra microorganisms, before treatment with streptomycin and infection with H37 Rv organisms.

CONCLUSION

1. The early dissemination of tubercle bacilli in the normal guinea pig is not affected by streptomycin therapy.

2. Although streptomycin limits the progression of tuberculous lesions in the guinea pig, lesions do progress despite adequate therapy, until acquired host-resistance develops, as shown by the development of skin hypersensitiveness.

CONCLUSIONES

La Estreptomicina en la Tuberculosis Experimental. III. Efecto sobre la Patogenia de la Tuberculosis Temprana en el Cobayo Infectado con Bacilos Tuberculosos H37 Rv Estreptomicinosensibles

1. La diseminación temprana de bacilos tuberculosos en el cobayo normal no es afectada por la estreptomicinoterapia.

2. Aunque la estreptomicina limita la agravación de las lesiones tuberculosas en el cobayo, las mismas continúan avanzando a pesar de una terapéutica adecuada hasta que se establece resistencia adquirida en el huésped, según revela la aparición de hispersensibilidad cutánea.

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STREPTOMYCIN IN EXPERIMENTAL TUBERCULOSIS[1, 2]

IV. Effect on the Pathogenesis of Early Tuberculosis in Guinea Pigs Infected with Streptomycin-resistant H37 Rv Tubercle Bacilli

PHILIP C. PRATT AND W. STEENKEN, JR.

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INTRODUCTION

In a previous paper (1), observations on the early course of experimental tuberculosis in streptomycin-treated and nontreated guinea pigs were presented. The infecting microorganisms used in that experiment were the H37 Rv streptomycin-sensitive tubercle bacilli (sensitive to 0.4 γ per cc. of streptomycin) when tested in Tween-albumin medium. In the present experiment, microorganisms (H37 Rv) were used which had been made resistant *in vitro* to 1,000 γ of streptomycin per cc. in Tween-albumin medium. Except for the change in the infecting microorganisms used, the conditions of the previous experiments were repeated as nearly as possible.

MATERIALS AND METHODS

A stock culture of H37 Rv streptomycin-sensitive tubercle bacilli was made resistant to the drug by repeated subculture in gradually increased concentrations of streptomycin until it had developed a resistance to more than 1,000 γ of streptomycin per cc. (2). This strain was subcultured in the presence of 1,000 γ of streptomycin per cc. of medium for six months.

Preparation of suspension of bacilli: An evenly dispersed 14 day growth of the highly resistant tubercle bacilli in Tween-albumin medium was diluted with physiological saline until it gave a reading of 17 in the Klett Summerson colorimeter. This produced a suspension containing approximately 20,000,000 tubercle bacilli per cc.

Infection: Twenty tuberculin-negative male guinea pigs, each averaging 450 Gm. in weight were used. Each animal was injected subcutaneously in the midline anterior to the symphysis pubis with 0.2 cc. of the suspension, or approximately 4,000,000 tubercle bacilli.

Therapy: Ten of the animals were used as infection controls and the other 10 were treated with streptomycin. Each of the treated animals was given an initial dose of 4,000 units of the drug forty-eight hours before infection, and thereafter 18,000 γ were administered daily in four divided doses: three doses of 4,000 each at 7 a.m., 12 noon, and 5:30 p.m., and an evening dose of 6,000 at 10:30 p.m. Treatment was continuous through the experiment.

Sacrifice periods: Two treated and 2 control animals were sacrificed 6, 12, 18, 38, and 66 days after infection. Studies by tissue subinoculation were not made on these animals since it was assumed that, on the basis of the previous experiment (1), the rate of spread of the resistant organisms in the control animals would be identical (or in exact ratio) to the spread of the sensitive organisms in the nontreated animals. The spread of the disease in the treated animals was obvious and did not necessitate subinoculation.

RESULTS

The essential gross and microscopic observations are summarized in the accompanying table which shows that lesions developed in the various organs at the

[1] From the Trudeau Laboratory of The Trudeau Foundation for the Clinical and Experimental Study of Pulmonary Disease, Trudeau, New York.

[2] This study was aided by a grant from the Division of Research Grants and Fellowships of the National Institute of Health, U. S. Public Health Service.

same intervals after infection and progressed at similar rates in the treated and in the control animals. In many instances, there appeared to be fewer Langhans' cells associated with the lesions in the treated animals than in the controls, but no gross and no other histological distinctions could be made.

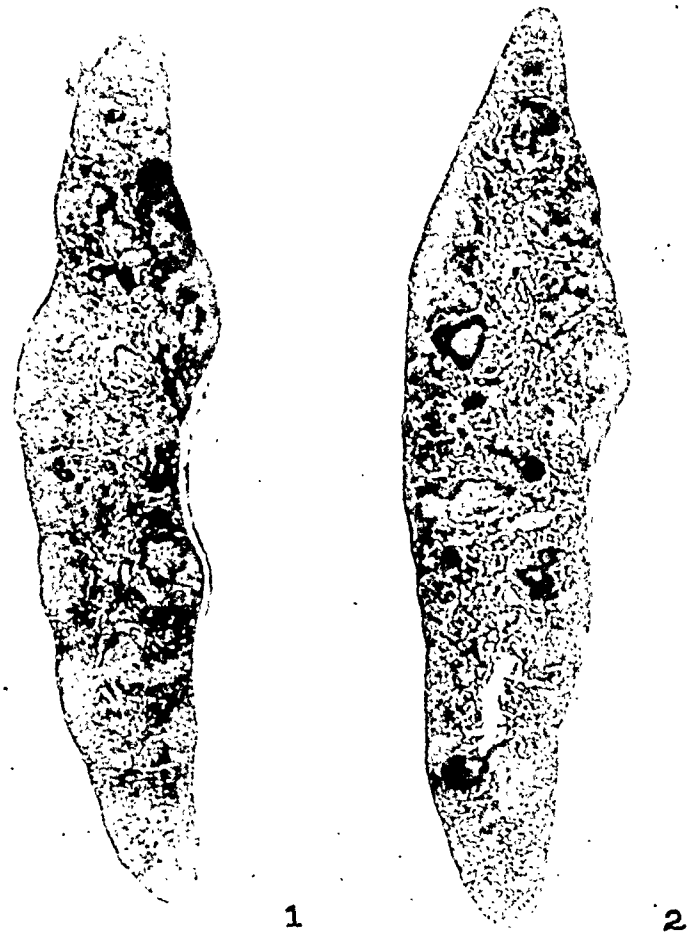

PLATE I

FIG. 1. Low power magnification of spleen from animal treated 66 days. Compare with figure 2. Note similar degree of enlargement. Tuberculous lesions are present in the malpighian bodies. ×10. (See Plate II, figure 1)

FIG. 2. Lower power magnification of spleen from control animal at 66 days. ×10.

COMMENT

The results of this experiment indicate clearly that guinea pigs infected with H37 Rv tubercle bacilli that were made highly resistant to streptomycin (1,000 γ or more) by *in vitro* adaptation do not respond to streptomycin therapy.

Other experiments to be reported from this laboratory by Steenken and Wolinsky (3) will show that guinea pigs infected with tubercle bacilli freshly isolated from patients under streptomycin treatment, and which have become

resistant to much lower concentrations than 1,000 γ of the drug per cc. of Tween-albumin medium, do not respond to streptomycin therapy. The same authors have also noted that a rough approximation of the relative number of drug-

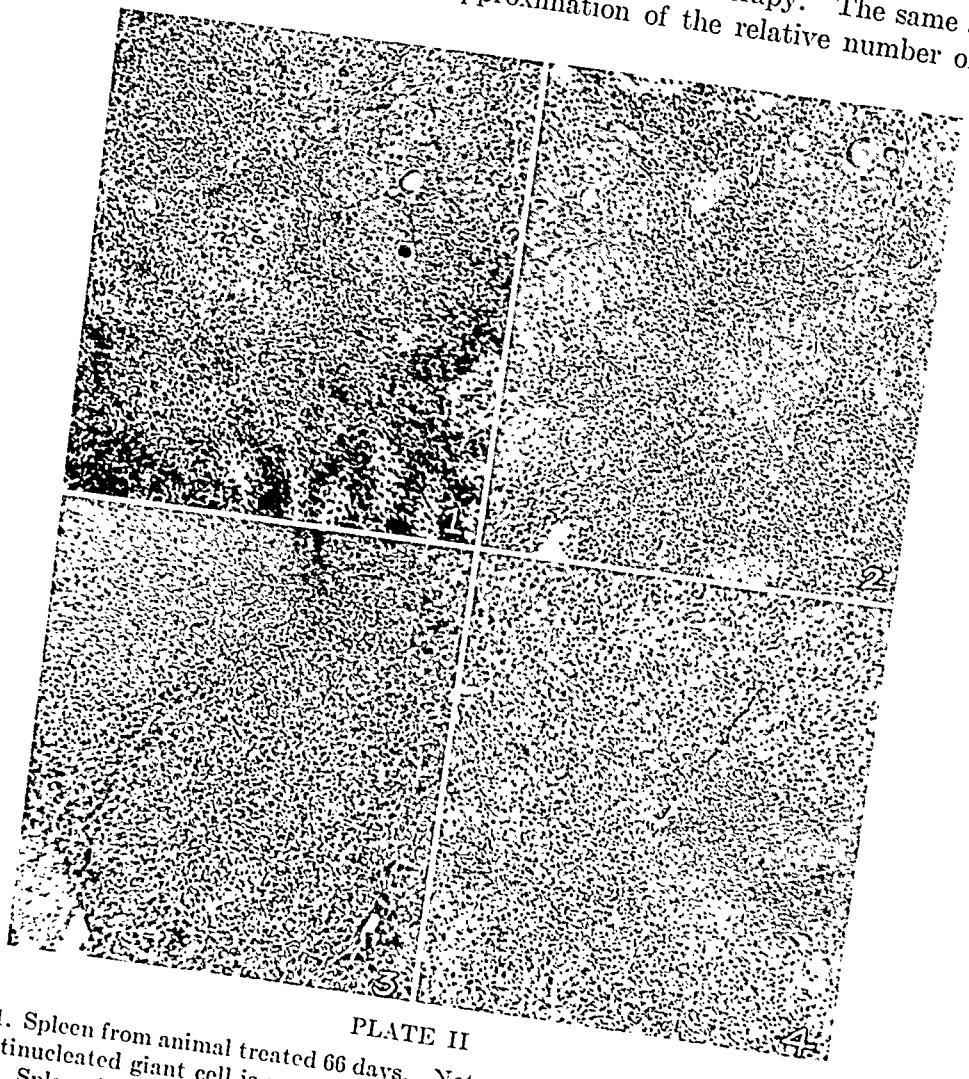

PLATE II

FIG. 1. Spleen from animal treated 66 days. Note unencapsulated epithelioid tubercles. One multinucleated giant cell is present in this field. ×120.

FIG. 2. Spleen from control animal 66 days. Note similarity to figure 3. Several multinucleated giant cells are present. ×120.

FIG. 3. Liver from animal treated 66 days. Note the presence of unencapsulated epithelioid tubercles. Necrosis is present in the center of the left upper quarter of the field. ×120.

FIG. 4. Liver from control animal at 66 days. Note similarity to figure 3. Necrosis is more obvious in this field. ×120.

resistant tubercle bacilli in a culture can be made by observing the length of the lag period before growth appears in a given concentration of the drug. It is possible that when resistance develops in a patient it is gradual and, therefore,

TABLE 1

Gross and Microscopic Pathological Findings in Streptomycin-treated and Nontreated Animals in Relation to Time after Infection

SACRIFICE PERIODS	TYPE OF PATHOLOGY	TREATED ANIMALS					CONTROL ANIMALS				
		Iliac node	Spleen	Lungs	Liver	Tracheobronchial node	Iliac node	Spleen	Lungs	Liver	Tracheobronchial node
6 days	Gross	Slightly enlarged	Slight* enlargement	0	0	0	Slightly enlarged	Slight* enlargement	0	0	0
	Microscopic	Macros in sinuses	0	0	0	0	Macros in sinuses	0	0	0	0
12 days	Gross	Enlarged with tubercles	Slight enlargement	0	0	Slight enlargement	Enlargement with tubercles	Slight enlargement	0	0	Slight enlargement
	Microscopic	Tubercles with necrosis	0	0	0	Macros* with sinuses	Tubercles with necrosis	One tubercle	One* tubercle	0 0	Macros in sinuses
18 days	Gross	Enlarged with caseous foci	Enlarged	Yellow flecks	Scattered tubercles*	Slight enlargement	Enlargement with caseous foci	Enlarged with tubercles*	Yellow flecks*	Scattered tubercles	Slight enlargement
	Microscopic	Epithelioid tubercle with caseation	Epithelioid tubercles	Epithelioid* tubercles + necrosis	Epithelioid tubercles	Epithelioid tubercles	Epithelioid tubercles + caseation	Epithelioid* tubercles + necrosis	Epithelioid tubercles	Epithelioid* tubercles	Epithelioid tubercles
38 days	Gross	Enlarged 4×	Enlarged 3×	Slight enlargement with gray flecks	Many tubercles	Enlarged 3×	Enlarged 4×	Enlarged 3×	Slight enlargement with gray flecks	Many tubercles	Enlarged 3×
	Microscopic	Epithelioid + caseous tubercles scarring	Epithelioid caseous tubercles	Epithelioid tubercles with caseation	Epithelioid + caseous tubercles	Epithelioid + caseous tubercles scarring	Epithelioid + caseous tubercles	Epithelioid + caseous tubercles	Epithelioid tubercles with caseation	Epithelioid + caseous tubercles	Epithelioid + caseous tubercles

* Noted in only one of the animals.

he may still respond to therapy for a certain period of time until his resistant organisms become predominant, and the patient no longer responds to the drug.

CONCLUSIONS

1. The early course of the disease in guinea pigs infected with H37 Rv tubercle bacilli resistant to 1,000 γ of streptomycin per cc. was not appreciably altered by streptomycin therapy.

2. Pathological lesions of tuberculosis under the conditions of this experiment were similar in extent in the treated and the control animals. The histological appearance of the lesions was similar in the two groups except that fewer Langhans' cells were seen in the treated animals.

CONCLUSIONES

La Estreptomicina en la Tuberculosis Experimental. IV. Efecto sobre la Patogenia de la Tuberculosis Temprana en los Cobayos Infectados con Bacilos Tuberculosos H37 Rv Estreptomicinorresistentes

1. La evolución temprana de la tuberculosis en cobayos infectados con bacilos tuberculosos H37 Rv resistentes a 1,000 γ de estreptomicina por cc. no fué alterada apreciablemente por la estreptomicinoterapia.

2. En las condiciones de este experimento, la patología de la tuberculosis es semejante en extensión en los animales tratados y los testigos. El aspecto histológico de las lesiones fué también semejante en los dos grupos, salvo por observarse menos células de Langhans en los animales tratados.

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TRANSITORY INFILTRATES IN THE LUNGS WITH EOSINOPHILIA (LOEFFLER'S SYNDROME) [1]

A Review With the Report of a Case

VICTOR F. WOOLF AND WILBUR J. GOULD

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REVIEW OF THE LITERATURE

The syndrome of pulmonary infiltration of brief duration associated with eosinophilia was described by W. Loeffler of Zürich in 1932 (1). By 1936 (2) he had accumulated 51 cases. Although most of this material came from chest clinics, in no instance was there any evidence of active pulmonary tuberculosis.

This group was of special interest because of the difficulties in differential diagnosis. The shadows observed in the lung fields were of many types, varied in distribution and were remarkable for their fleeting character. In some patients the process was migratory. Another distinguishing characteristic was the presence of an eosinophilia. In more than half of these individuals, the eosinophils ranged between 10 and 30 per cent. The highest percentage of eosinophils observed was 66. There was no strict parallel between the size of the infiltrate and the accompanying eosinophilia. The maximum number of eosinophils was found at the time when the pulmonary shadows had begun to resolve. Very often, the eosinophilia persisted for a long time after the clearing of the pulmonary process in the roentgenogram. The symptomatology also bore no constant relation to the extent of the physical and roentgenological findings.

Fourteen of Loeffler's 51 cases were entirely symptom-free. In these patients the pulmonary shadows were discovered during the course of routine fluoroscopy or roentgenographic surveys. The majority of the patients complained chiefly of fatigue and of respiratory symptoms of varying severity. The condition was encountered most frequently between July and August. Fully two-thirds of the patients were males. Two instances were found in members of the same family. All the cases followed a benign course regardless of the therapy employed. The individuals observed in Loeffler's series were of different age groups; one-third were small children. Thirteen had negative skin reactions to tuberculin, while the remaining 37 were tuberculin-positive.

Since Loeffler's observations, the literature on the subject has grown considerably. Reports have come from all parts of the world. Virtually all authors now agree that the condition is an allergic reaction occurring in the lungs of a sensitized individual. Most of the patients are found to have an allergic background and to have had other manifestations of allergy. The number and variety of conditions in which Loeffler's syndrome has appeared is constantly increasing and may be limitless. Cases have been reported in the course of, or in association with, various bacterial infections, parasitic infestations, asthma, pollen sensitivity, and other allergic states, erythema nodosum, periarteritis no-

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dosa, drug therapy, and exposure to physical agents. The syndrome has appeared in groups (21), in "epidemics" (22) and sporadically. The latter occurrence is more frequent.

In some instances the process in the lungs and the eosinophilia have recurred in the same individual. In others they have followed a more prolonged course of from several weeks to several months and have been associated with severe manifestations in other organs. Harkavy (3) reported a series of 16 cases of bronchial asthma with diffuse vascular involvement, almost all of which showed Loeffler's Syndrome on one or more occasions over a period of several years.

Because of the apparently benign course in most persons affected, very few studies of the tissues have been made in the living patient. The sputum (3, 14), pleural fluid (3, 14), peritoneal fluid (3), lung puncture (14), and muscle biopsy (3) material have shown a preponderance of eosinophils when examined.

Von Meyenburg (4) described the postmortem findings in 4 cases of accidental death, one of which followed traumatic tetanus. These are believed to show Loeffler's Syndrome. In 3 there were areas of focal pneumonia; in 2 there were bronchopneumonias in which the exudate was largely eosinophilic; and one showed evidence of beginning organization of the exudate in a few places. Two showed eosinophilic bronchitis and bronchiolitis. Perivascular and interstitial lesions were also described. In one instance a necrotic focus was found in a pneumonic area. Some areas of thrombophlebitis were found in the septal veins, none of which was completely obstructed. Eosinophilic infiltration was found in the other organs, particularly in the bone marrow and in the liver. Charcot-Leyden crystals were found especially after the tissues had lain in the refrigerator.

Von Meyenburg felt that the anatomic basis of Loeffler's Syndrome had been demonstrated. He did not feel that the fleeting nature of the lesions had been proved, at least in one of the cases, because of the nature of the involvement of the interstitial tissues and blood vessels. This would correspond with the cases of longer duration described by several authors (5, 6, 7, 8, 9, 10, 11, 12). He concluded that the lung lesions of Loeffler's disease "are really exudative foci, eosinophilic pneumonias."

He also described a nonspecific eosinophilic epididymitis in an operative specimen removed from a patient who had a transient lower lobe shadow associated with eosinophilia. From this observation, as well as from the findings in the other patients, he concluded that eosinophilic infiltrates occur simultaneously in other organs. Of the 6 deaths in Harkavy's (3) series, 4 came to postmortem examination. The lungs showed congestion, edema, and infiltration of the interalveolar septa with eosinophils, neutrophils, and lymphocytes as well as edema. There were also noted atelectasis, emphysema, and infarction, depending on the preceding course and complications. The bronchi exhibited the changes commonly seen in asthma. Most interesting were the lesions in the blood vessels varying from simple thickening of small vessels involving the intima to an acute necrotizing arteritis with periarterial eosinophilic infiltration and endarteritis obliterans. In the pleura there were sclerosis of small vessels

and inflammatory exudate with eosinophilia. The other organs showed similar vascular lesions. In 2 of these autopsies a pathological diagnosis of periarteritis nodosa was made.

Bagenstoss, Bayley, and Lindberg (13) report on the postmortem pulmonary findings in a case of Loeffler's Syndrome. The lungs showed scattered tubercle-like granulomata. In some areas there was organization of the alveolar exudate. A more recent acute inflammatory process was found in the lower lobes with a predominance of eosinophils in the exudate. The bronchioles contained mucus and desquamated epithelial cells. Their walls were infiltrated with eosinophils and plasma cells. Perivascular inflammatory lesions with thickening and occasional necrosis of the vessels walls were also present.

Von Meyenburg (4), Nagel (14), and Herbut and Kinsey (23) cited experimental work (15, 16, 17) in which the various clinical and pathological findings observed in individuals with eosinophilic lung infiltrates have been reproduced. Hansen-Pruss and Goodman (18), Bagenstoss, Bayley and Lindberg (13) and Harkavy (3) mention the role of the vascular system in producing the findings in their patients. They refer to the work of other authors wherein similar vascular lesions are found to be on an allergic basis. Alpher (19) regards the occurrence of eosinophilic infiltrates in other organs as postulating a hematogenous pathway for the "intrinsic type" of case and he raises the possibility that Loeffler's Syndrome may be due to viral infection in an essentially allergic person. He also regards the interstitial tissue as the shock organ whereas Hansen-Pruss and Goodman (18) offer the view that in the course of repeated respiratory infections there is possible sensitization of the bronchial tree by bacteria with allergic response in the form of a pneumonic type of reaction to reinfection with the sensitizing antigen or with some other antigen.

The importance of the syndrome lies in the problem of differential diagnosis which it presents. Some cases have been mistaken for tuberculosis, as with the patient in the present report, some for lung abscess or infarcts, and others for pneumonia of various kinds.

The types of shadows seen on the chest roentgenograms, according to Loeffler (2) are:

1. Large, more or less irregular shadows, unilateral or bilateral.
2. Small round foci.
3. Multicentric unilateral or bilateral infiltration.
4. Lobar shadows.
5. Infiltrates simulating "secondary tuberculous infection."
6. Cases with pleural involvement and a very small fleeting effusion.

In addition to the varied shadows noted by others, Hennell and Sussman (15) describe:

narrow, plate-like homogenous densities. . . . .extending obliquely caudad and laterally. Often they are symmetrical in the two lungs. They also resolve completely. Whether they represent localized exudations in the lung or pleura is impossible to state, but they seem to be unique to this disease. There is no predilection for upper or lower lobes, but a fair degree of symmetry on the two sides is the rule.

The physical signs are variable depending upon the extent of involvement. The following case is reported as an instance of Loeffler's Syndrome.

CASE REPORT

S. P., a 44 year old white housewife and factory worker, was a known asthmatic for about six years. Following an episode of bronchopneumonia in 1939, the asthmatic attacks first appeared and increased in severity. There was no familial history of allergy. The patient had been studied in the Allergy Clinic and was found to be slightly sensitive to dust, cat dander, dog dander, goat hair, flaxseed, cotton seed, tobacco, and white potato. The last asthmatic attack followed a cold during August, 1945. For three weeks prior to the first visit to the Medical Clinic on September 13, 1945, the patient had noted fatigue, dyspnea on effort, irritability, nervousness, loss of appetite and a 10 pound loss of weight.

A chest roentgenogram taken September 13, 1945 (figure 1) revealed heavy infiltration throughout the upper portions of both lung fields, more prominent on the left side. There was some patchy infiltration in the lateral portion of the right lung opposite the hilum and near the diaphragm. The cardiac shadow showed slight generalized enlargement.

A blood count performed on September 13, 1945 showed: erythrocytes 4,400,000, hemoglobin 13 Gm. per 100 cc., leukocytes 12,300 per cu. mm., neutrophils 62 per cent, lymphocytes 24 per cent, eosinophils 14 per cent. The urine was normal and the blood pressure in mm. of mercury was 110 systolic and 70 diastolic. The patient was referred to the Pulmonary Clinic where examination on September 24, 1945 revealed dullness at both apices. No rales were heard. Hospitalization for tuberculosis was advised. She was admitted to another hospital on September 26, 1945 with the diagnosis of bilateral pulmonary tuberculosis, far advanced. On admission the blood pressure was 170 systolic and 100 diastolic. Otherwise there was no change in the physical findings.

A roentgenogram of the chest obtained on September 28, 1945 (figure 2) showed dense infiltration diffusely distributed throughout the left lung. The upper portions of the right lung field revealed similar, though less extensive, infiltrates. A subsequent roentgenogram taken on December 21, 1945 (figure 3) showed that the pulmonary infiltrates had resorbed completely. There was marked enlargement of the heart when compared with the cardiac silhouette in the previous film.

The treatment consisted of bed rest with symptomatic therapy. Examinations of five sputum concentrates were negative for acid-fast bacilli. The sedimentation rate was 32 mm. per hour. During her stay in the hospital the patient gained ten pounds in weight. The final diagnosis on discharge was nontuberculous pulmonary disease, probably pneumonia.

FIG. 1. Chest roentgenogram taken on September 13, 1945 shows an exudative infiltrate involving the upper portions of both lungs, but more dense and extensive on the left. The lateral portions of the right first to fourth anterior interspaces and of the base are also involved.

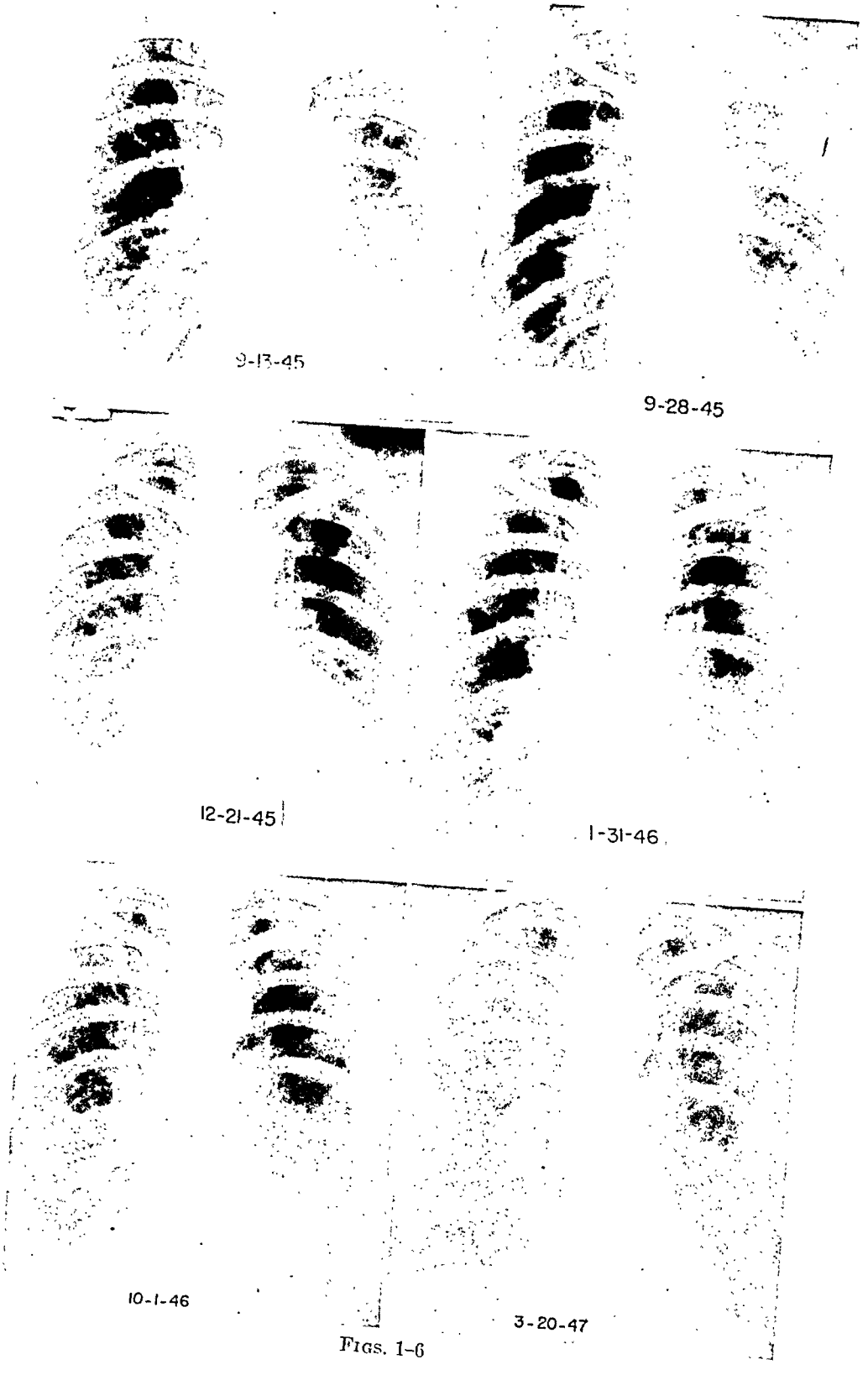
FIG. 2. Chest roentgenogram taken on September 28, 1945 shows some clearing in the right upper lung field. There has been some increase in the shadows previously noted at the periphery and base of the right lung. The left shows shadows of varying size and density throughout.

FIG. 3. Chest roentgenogram taken on December 21, 1945. The lung fields are clear except for some accentuation of the markings and fine nodular mottling in the lower portions. The heart is considerably enlarged.

FIG. 4. Chest roentgenogram taken on January 31, 1946. The lung fields are clearer; the markings somewhat accentuated. The heart is smaller.

FIG. 5. Chest roentgenogram taken on October 1, 1946. The lung fields are clear. The heart is smaller.

FIG. 6. Chest roentgenogram taken on March 20, 1947. Lung markings are accentuated. The heart is unchanged.



FIGS. 1–6

The patient returned to the Pulmonary Clinic on February 4, 1946 with complaints of nervousness and irritability. Physical examination revealed clear lungs. The blood pressure in mm. of mercury was 144 systolic and 92 diastolic.

A roentgenogram taken on January 31, 1946 (figure 4) again showed complete clearing in both lung fields.

The Mantoux test was positive to 0.01 mg. of Old Tuberculin. The stool contained no ova or parasites. A blood count on March 22, 1946 showed erythrocytes 4,600,000, hemoglobin 14.2 Gm. per 100 cc., leukocytes 11,500 per cu. mm. with 66 per cent neutrophils, 24 per cent lymphocytes, 7 per cent eosinophils, and 3 per cent basophils.

A diagnosis of Loeffler's Syndrome was made.

The blood pressure ranged from 160 systolic and 100 diastolic on January 19, 1946 to 142 systolic and 88 diastolic on March 25, 1946. An electrocardiogram taken on February 25, 1946 showed sinus tachycardia, left axis deviation, low diphasic T waves in leads I and II, and a small R wave in lead III.

The patient remained well until April 1946, when she was hospitalized because of cough, dyspnea and wheezing of two weeks' duration. The patient was believed to have a respiratory infection and was given penicillin intramuscularly. Epinephrin, aminophyllin and "benadryl" were used to control the wheezing which disappeared after a few days. She left the hospital feeling well The blood pressure on admission had been recorded as 110 systolic and 80 diastolic but fluctuated between 142 and 110 systolic and 90 and 60 diastolic. The leukocyte count was 13,800 per cu. mm. with 78 per cent polymorphonuclears, and 22 per cent lymphocytes. Two specimens of sputum were negative for tubercle bacilli, and examination of the stool showed no ova or parasites. No roentgenogram was obtained on this admission.

Subsequently the patient had several less severe asthmatic attacks and was treated in the Allergy Clinic with aminophyllin and injections of vaccine and bacterial extract. On April 1, 1947 her total leukocyte count was 7,000 per cu. mm. with 56 per cent neutrophils, 28 per cent lymphocytes and 16 per cent eosinophils. The sputum was negative for tubercle bacilli and contained numerous eosinophils.

On October 1, 1946 (figure 5) a roentgenogram showed somewhat less prominent lung markings and some diminution in the heart size. The latter was slightly smaller than in the earliest film and the apex was more clearly defined. The most recent film take March 20, 1947 (figure 6) showed the markings somewhat more accentuated than in the two preceding films, but no increase in the heart size.

DISCUSSION

The illness of this patient, presented as showing Loeffler's Syndrome, seems to resemble those instances of longer duration and greater complexity with involvement of extrapulmonary organs which were described since Loeffler's original observations.

The changes in heart size and the elevated blood pressure in this patient suggest the possibility of widespread eosinophilic infiltration in the heart and other organs. Unfortunately, no electrocardiograms were taken during the period of greatest cardiac enlargement. No evidence of renal damage was found.

Some of Harkavy's (3) patients showed electrocardiographic changes, pericarditis, and signs of cardiac enlargement or failure. In the nonfatal cases the changes were reversible, but recurred with subsequent asthmatic attacks. All of his 6 fatal cases showed clinical or pathological signs of adhesive pericarditis. Three had constrictive pericarditis and 4 had polyserositis. One of Karan and Singer's (11) cases showed temporary cardiac enlargement, chiefly of the right ventricle. This was attributed to right ventricular strain and regressed with the

resorption of the pulmonary shadows. It will be interesting to follow this patient's course in the light of Harkavy's findings.

Experience with this patient serves to illustrate the differential diagnostic problem. In this instance pulmonary tuberculosis was suspected because of the symptoms and roentgen picture. The patient actually spent three months under observation for pulmonary tuberculosis.

Because the symptomatology may simulate both acute and chronic pulmonary diseases and since the roentgenographic findings are extremely varied, it is important that the patient be studied from the clinical as well as from the laboratory standpoints. Loeffler's Syndrome should be suspected if the blood count repeated at intervals of several days to a week shows eosinophilia. A careful history may disclose repeated respiratory infections superimposed on an allergic background such as asthmatic attacks, hay fever, migraine, et cetera.

Roentgenograms taken at intervals of one week will usually show rapid clearing or migration of the pulmonary shadows. The presence of the oblique shadows described by Hennell and Sussman (20) may be helpful. The sputum should be examined for tubercle bacilli. The absence of acid-fast organisms, especially when large lesions exhibiting lobar distribution and rarified areas suggesting cavitation are present, would tend to exclude a diagnosis of pulmonary tuberculosis. Pneumococcus pneumonia can be excluded by the failure to find pneumococci of the fixed types in the sputum. The use of the cold agglutination test may be valuable in ruling out the atypical pneumonias. The absence of foul or purulent sputum and the rapid resolution help to separate these cases from lung abscess and suppurative pneumonia. Observation, bronchoscopy, and serial roentgenograms will differentiate Loeffler's Syndrome from carcinoma of the lung. Pulmonary infarct can be distinguished by the course and the absence of conditions with which it is usually associated.

Because of the frequent association of this syndrome with chronic infections, especially by protozoa, stool examinations, cutaneous tests, and serological procedures may be of great value in detecting unsuspected infections whose causative organisms may act as sensitizing and provocative antigens. A thorough allergic study with various antigens may be advisable. As this condition may be but one manifestation of a serious underlying disease such as periarteritis nodosa, especially in the more protracted or recurrent cases, appropriate steps should be taken to exclude this possibility.

SUMMARY

A case of Loeffler's Syndrome in a 44 year old woman is presented. Transitory cardiac enlargement and mild hypertension were also observed.

SUMARIO

Infiltraciones Transitorias en los Pulmones con Eosinofilia (Síndrome de Loeffler)

Este caso de síndrome de Loeffler en una mujer de 44 años coexistió con hipertrofia cardíaca pasajera y leve hipertensión. El síndrome de Loeffler pasa por ser un fenómeno alérgico exudativo en los pulmones.

Acknowledgment

The authors wish to acknowledge the courtesy of Dr. F. J. McCarthy in allowing them to use the records of this case at St. Joseph's Hospital, Bronx, New York, N. Y.

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TUBERCULOUS ESOPHAGO-CUTANEOUS FISTULAE TREATED WITH STREPTOMYCIN AND GASTROSTOMY[1]

Report of a Case

JOHN F. LAWSON[2]

(Received for publication June 28, 1948)

This case is presented primarily as an unusual form of tuberculosis and, secondly, because one phase of the disease responded to streptomycin.

CASE REPORT

History: A 28 year old, white, single female was admitted to North Carolina Sanatorium on October 7, 1947 with the following history. She enjoyed good health and normal development until 1924, when at the age of five she was exposed to a known case of tuberculosis. Shortly after this she is said to have developed measles and an upper respiratory infection, followed several weeks later by pain in the thoracic spine. A diagnosis of tuberculosis of the spine was made and a body cast was applied and worn for one year. In spite of this, she had a marked kyphosis over the lower cervical and upper thoracic vertebrae, which was painless and which allowed normal physical activity for the next twenty years.

In June, 1944 she noted a hard swelling the size of a hen's egg on the left side of the neck associated with generalized malaise and fever. Tuberculous abscess of a cervical lymph node was diagnosed and the swelling was later incised and drained with an indwelling tube. This area never healed, and eleven months later while drinking a beverage the patient discovered that the latter had seeped through the sinus and had wet the exposed surface of the neck. Pain reappeared in the spine and shortly thereafter, in 1945, a mass was noted on the right side of the neck. This, in due time, followed a course similar to that on the left. Both sinuses have continued to drain purulent material intermittently since 1945. Body cast was applied at that time and the patient continued to wear this. However, the pain in her spine persisted and the kyphosis became more marked.

Physical examination: Physical examination at the time of admission revealed a small, poorly developed and poorly nourished, white female who appeared chronically ill. Her temperature was 99° F., pulse 120 per minute and the respirations were 18 per minute. The blood pressure in mm. of mercury was 144 systolic and 100 diastolic. There was generalized wasting of the subcutaneous tissues. Examination of the head revealed nothing abnormal. The neck was thrust forward as a result of the spinal deformity. Bilateral, nontender, anterior cervical lymph nodes were felt. At the base of the neck on each side just anterior to the trapezius muscle were scarred areas the size of a half-dollar with small fistulous openings from which purulent material could be expressed. Fluids taken by mouth appeared promptly at these openings. There was marked kyphosis involving the lower cervical and upper thoracic vertebrae with a nontender gibbus involving the third and fourth thoracic vertebrae, and accompanying thoracic deformity of marked degree. The heart and lungs were normal on percussion and auscultation.

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Neurological examination was normal. The remainder of the physical examination was noncontributory.

Laboratory examination: Studies of the blood revealed no abnormalities except for 12,750 leukocytes per cu. mm. with 85 per cent neutrophils, 14 per cent small lymphocytes,

FIG. 1

FIG. 2

FIG. 3

FIG. 1. Chest roentgenogram demonstrating marked destruction of thoracic vertebrae.
FIG. 2. Lateral roentgenogram showing spinal deformity.
FIG. 3. Lipiodol was injected into both sinuses and could be traced by fluoroscopy into the esophagus and stomach. The saccular area in the region of the esophagus is not demonstrated in this roentgenogram.

and 1 per cent eosinophils. A serologic test for syphilis (Kline) was negative and the urinalysis was normal.

Roentgenograms of the chest showed no evidence of active pulmonary tuberculosis. There was definite widening of the superior mediastinum, however, particularly on the left. Roentgenograms of the spine revealed marked destruction of the dorsal vertebras as shown in figures 1 and 2. Lipiodol was injected into each sinus and this could be

traced by fluoroscopy into a saccular area that was continuous with the esophagus and stomach. This is shown in part by figure 3. The saccular area retained the opaque material only transiently. Sputum, examined repeatedly by concentration and culture, was negative for tubercle bacilli. Cultures from the draining fistulae revealed tubercle bacilli.

FIG. 4

FIG. 5

FIG. 4. Photograph representing topographical deformity of spine. Dressing on abdomen covers site of gastrostomy incision. Supraclavicular scar is visible but is shown in higher magnification in figure 5.

FIG. 5. Enlargement to show healed scar in supraclavicular region.

Course in hospital: A diagnosis was made of tuberculous spondylitis complicated by formation of mediastinal abscess, bilateral esophago-cutaneous fistulae, and bilateral tuberculous cervical lymphadenitis. Because of the results reported (1, 2) from the use of streptomycin in the treatment of draining tuberculous sinuses, this drug was used in this case. Esophagoscopy was attempted as a preliminary to chemotherapy, but was unsuccessful because of the spinal deformity. Intramuscular streptomycin, 1 Gm. a day in 3 doses, was begun on November 27, 1947.

On November 28, 1947 gastrostomy was performed in order to put the esophagus at rest during streptomycin therapy. The patient was fed via the gastrostomy and received nothing by mouth for seventy-two days. After one month of treatment the fistula on the left side closed, followed on the seventh week by closure of the fistula on the right. On February 6, 1948 the patient was given a soft, bland diet by mouth and on February 19 the fistula on the left side reopened. Streptomycin was discontinued on February 20 after the patient had received 86 Gm. in eighty-six days, the esophagus remaining free from food for seventy-two days during this period. The fistula on the left drained for three weeks and closed spontaneously and neither fistula has drained for the past three

FIG. 6. Barium swallow, three months after streptomycin was discontinued, demonstrating the distorted esophagus. No fistulous tracts are seen.

months.[3] After two previously unsuccessful attempts, the gastrostomy was closed on May 11, 1948.

At present the patient is taking a regular diet, eating well, and gaining weight. There are no palpable cervical lymph nodes. Figure 4 presents a photograph taken May 8, 1948 showing the deformity. Figure 5 illustrates the healed site of the previous fistulous opening on the left, ten weeks after streptomycin therapy was discontinued. The right side is not shown, but is identical with the left. Figure 6 presents the results of a barium swallow three months after streptomycin therapy and demonstrates the distorted esophagus which is deviated to

[3] Since this report was accepted for publication, the writer has been informed that one of the fistulae has reopened and continued to drain. No further streptomycin has been given.

the left in its upper portion. No fistulous tracts are seen. However, there is no knowledge of the condition of the presumed underlying mediastinal abscess.

The possibility that one or both fistulae were connected with the distal pharynx rather than the esophagus cannot be excluded with absolute certainty. The esophageal site is much more likely, however, because all of the pharynx, except the region of the junction between inferior constrictor muscle and esophagus, was observed under direct vision during the attempt at esophagoscopy.

SUMMARY

A case of tuberculous esophago-cutaneous fistulae treated with streptomycin is presented. Eighty-six grams of streptomycin were given over a period of eighty-six days. A gastrostomy was performed and for seventy-two days during the period of chemotherapy the patient was fed exclusively by tube. Both fistulae have remained closed for more than three months.[3]

SUMARIO

Fístulas Esófago-Cutáneas Tuberculosas Tratadas con la Estreptomicina y la Gastrostomía

El caso descrito es de fístulas esófagocutáneas tuberculosas tratadas con la estreptomicina, administrándose 86 Gm. de la droga durante un período de ochenta y seis días. Ejecutóse una gastrostomía y durante setenta y dos días de dicho período de quimioterapia el enfermo fué alimentado exclusivamente con sonda. Ambas fístulas han permanecido cerradas por más de tres meses.

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SKIN TUBERCULIN REACTION FOR THE ASSAY OF TUBERCULIN IN GUINEA PIGS[1]

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INTRODUCTION

Since the initial discovery by Koch that tuberculin could be a useful aid in the diagnosis of tuberculosis, its use has grown until it now occupies a position of universal acceptance. Many of the problems that have arisen in the use of tuberculin have resulted from a lack of knowledge of its chemical and immunological characteristics. Attempts to correlate various separately conducted studies on tuberculin skin sensitivity have not always been entirely successful. This is easily understood when consideration is taken of the many and varied methods and steps in the preparation of tuberculins. The use of various nonsynthetic and synthetic media, concentration of filtrate to standard volume without regard to its activity, and various methods of partial purification, all contribute to the confused picture found in correlation studies.

The development by Seibert (1) of a highly purified tuberculin, Purified Protein Derivative (PPD), has greatly reduced the nonspecificity of tuberculin reactions and furnished an excellent material to use as a tool for the comparison of various tuberculin preparations. Attempts at standardization of tuberculins, however, have shown both the unreliability of present methods and a great variation among tuberculins from various sources. Savage (2), Sweany (3), Seideman (4), and others have compared tuberculins from various sources and found considerable variation among products. Early attempts were made by several to establish a quantitative biological test. The determination of a minimal lethal dose of tuberculin injected subcutaneously into a tuberculous guinea pig was one of the earliest methods (5). Long (6) employed a method which determined the minimal amount of tuberculin which when injected into the testis of a tuberculous guinea pig would cause complete inhibition of spermatogenesis. Both of these methods proved impractical because results varied too greatly with the degree of tuberculin sensitivity. Bunney and Gottschall (7) and Clark and Follin (8) have presented semi-quantitative methods based upon comparative skin reactions in guinea pigs.

In view of the importance of the dosage of tuberculin in diagnostic, therapeutic, and prophylactic procedures, a standard method is required both for the standardization of tuberculin and for the clinical assessment of the tuberculin reaction to which standardized statistical procedures can be applied with consistent and reproducible results that can be reduced to a formula. The work to be reported in this paper was planned in an attempt to achieve these objectives.

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METHODS

White guinea pigs weighing between 375 and 400 Gm. were inoculated intraperitoneally with virulent *M. tuberculosis* following a procedure used routinely in this laboratory. Four to five weeks later, the skin on the back and sides of these animals was depilated by clipping and by rubbing with barium sulfide paste. Three or four graded doses of a tuberculin preparation (PPD) were injected in 0.1 ml. volumes intradermally on each side of the body. In case an unknown was compared with a standard, they were injected alternately on the same side. The dosages employed were those recommended for skin testing in humans.

The skin reaction was determined by measuring the diameter of the erythema evolved twenty-four and forty-eight hours following inoculation. The intensity of the reaction reached a maximum around twenty-four hours. When the reacting area was not exactly circular in shape, two diameters (maximum and minimum) were measured and the mean diameter was used in calculating the dose-response relationship.

In some experiments, the tuberculous pigs which had once been employed for skin testing were used again two to four weeks later. The results of this practice have been carefully checked and no tuberculin desensitization or lack of response has been found as long as the animals remained in sound physical condition.

RESULTS AND DISCUSSION

The data will be presented in two parts: (A) An analysis of the dose-response characteristics of the skin tuberculin reaction, and (B) an assay method for tuberculin preparations.

A. Skin Reaction to Dose of Tuberculin

The skin reaction to dose of tuberculin follows a linear relationship when the value of response (Y) and the dosage (X) are expressed in logarithms (figure 1).

$$\log Y = \log a + b \log X \qquad \text{Equation 1}$$

$$a,\ b = \text{constants; or}$$

$$Y = aX^b \qquad \text{Equation 2}$$

Obviously there is the same relationship whether the reaction is expressed by the diameter (D) of the erythema, its area—$\pi\left(\frac{D}{2}\right)^2$, its volume—$4/3\,\pi\left(\frac{D}{2}\right)^3$ or any other power of D (since $\log Y^c = C \log Y$).

The equation holds true for readings taken either at twenty-four or forty-eight hours after intradermal injection of tuberculin (figure 1). The twenty-four hour reading is favored, because the reaction is maximal at this time; furthermore, necrosis occurs at forty-eight hours with large doses of tuberculin.

Using area as a measure of response twenty-four hours after inoculation, and the value 20 for 0.00002 mg. of PPD, the equation 3

$$Y\,(D^2) = 17.0\,X^{0.349}$$

was found to fit the data in table 1. In deriving equation 3, there were 360 determinations (4 from each pig) which were obtained in 4 experiments (22 to 24 animals each). The estimates of the constants of equation 3 thus represent those for a large population. In early calculations (as for equation 3), the diameters

of the erythema at right angles were multiplied to obtain the area. The average diameter was used in later computations. The difference in values, as obtained

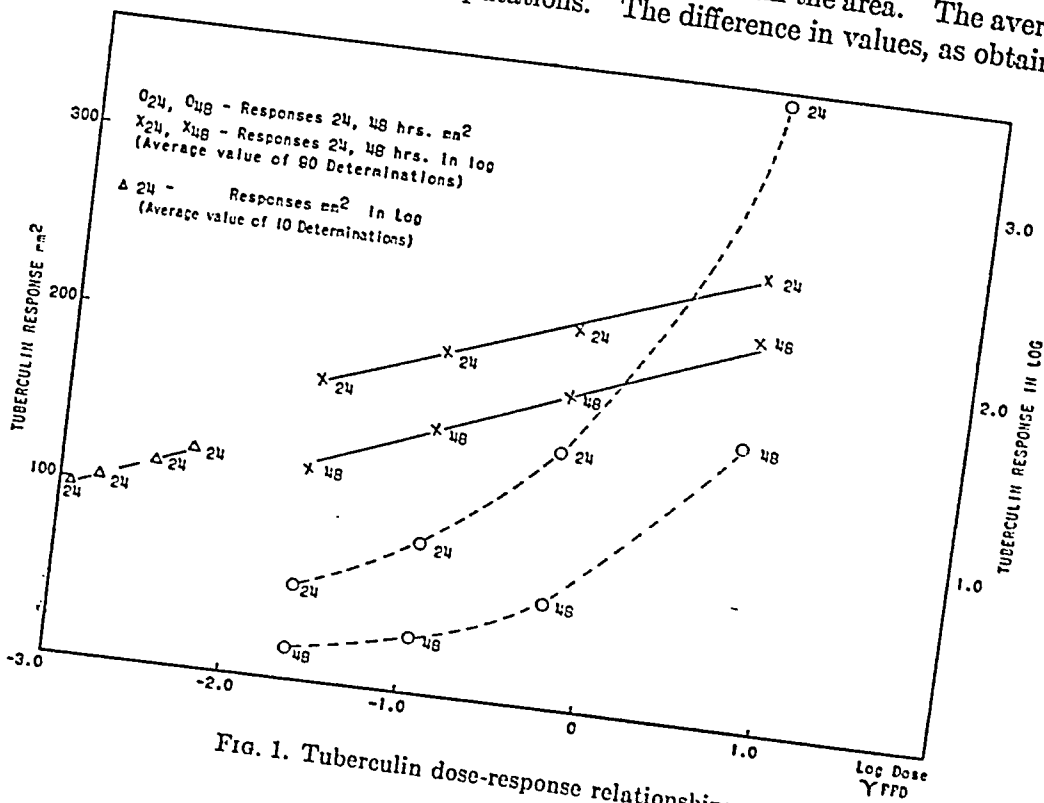

FIG. 1. Tuberculin dose-response relationships

TABLE 1

Tuberculin Skin Reaction

EXPERIMENT NUMBER	NUMBER OF PIGS	DOSE OF PPD/*SKIN REACTION IN MM.²							
		0.00002 mg.		0.0001 mg.		0.0005 mg.		0.005 mg.	
		24 hrs.	48 hrs.	24 hrs.	48 hrs.	24 hrs.	48 hrs.	24 hrs.	48 hrs.
1	22	64.9	23.2	97.0	31.5	154.7	61.7	418.8	174.7
2	22	51.1	19.9	89.9	40.5	152.2	64.4	351.2	187.1
3	22	46.6	15.5	81.3	30.2	141.7	53.7	333.6	137.5
4	24	38.4	8.1	62.6	20.0	115.0	46.4	301.2	136.4
Average value of 90 pigs.....		50.3	16.7	82.7	30.6	140.9	56.6	351.2	158.9

* = Average values of the number of guinea pigs in each experiment.

Y = Skin reaction at 24 hours. $Y = 13.5\ X^{0.349}$,

X = Dose of PPD (a value 20 for 0.00002 mg.)

in two ways, is very slight and is not significant statistically. The exponent 0.349, the slope of the regression line (equation 1), was computed by the method

of least squares. It has a standard error of ±0.025. Equation 3 may therefore be written as

$$Y(\text{in}) = 12.0\,X^{1/6} \text{ or } Y(\text{in}) = \sqrt{17.0}\,X^{1/3}$$

In other words, a straight line may be obtained by plotting the response in mm² against the cube root of the dosage of tuberculin or the response in diameter

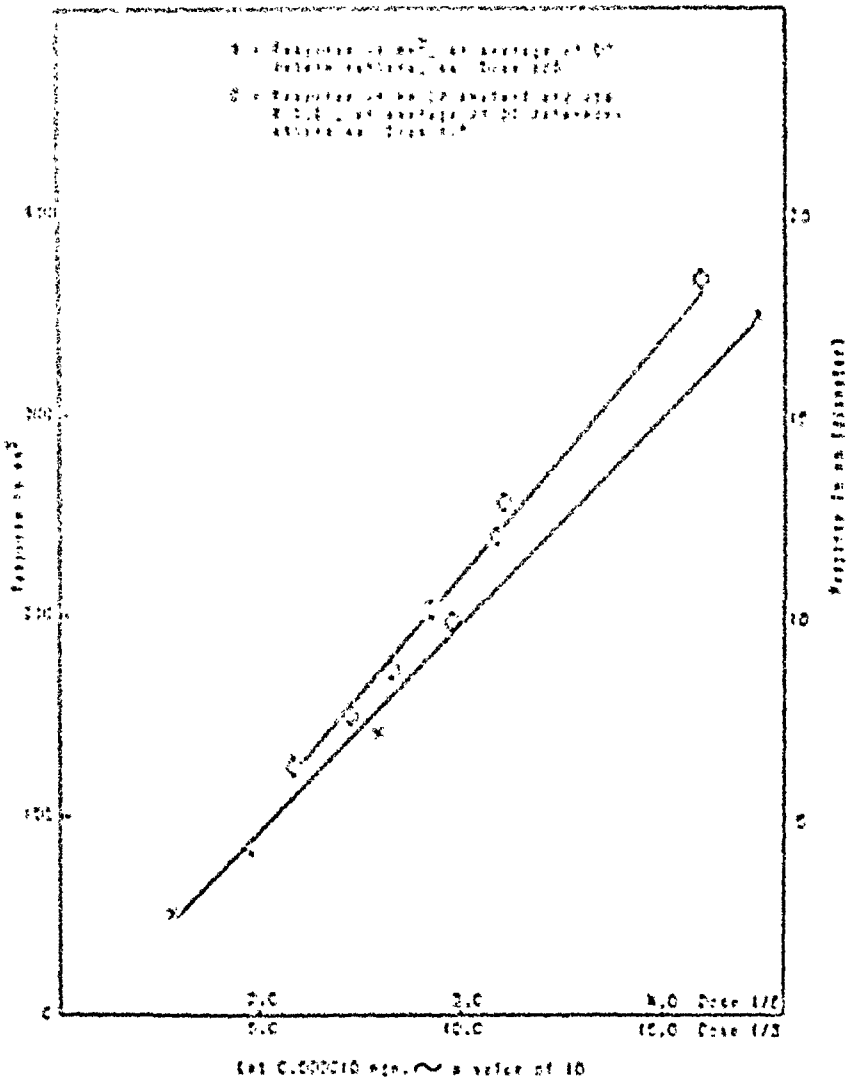

FIG. 2. Tuberculin dose-response relationship

against the 1/6 power of dosage. This is shown in figure 2. The two lines were drawn from data obtained in two separate experiments. One was plotted with the same data used to derive equation 3. The standard error of the mean indicates approximately the variation of the slope of the line. Fairly accurate estimation of slope may be obtained graphically with a much smaller number of

determinations as shown by graphs in figure 1. The question as to how many determinations are required in an accurate biological assay of tuberculin is discussed in the following section on assay method.

B. *Assay of Tuberculin*

From the dose-response relationship given above, an approximate estimation of the activity of an unknown tuberculin in reference to that of a standard tuberculin may be determined as follows:

Let X_{s1}, X_{s2}, X_u and Y_{s1}, Y_{s2} and Y_u be the doses and skin reactions in diameter (or area) of the erythema produced by the standard and the unknown tuberculin preparation respectively:

$$Y_{s1} = aX_{s1}^b, \qquad Y_{s2} = aX_{s2}^b, \qquad Y_u = aX_u^b$$

$$\frac{Y_{s2}}{Y_{s1}} = \frac{aX_{s2}^b}{aX_{s1}^b} = \left(\frac{X_{s2}}{X_{s1}}\right)^b, \qquad \text{or} \quad \log\frac{Y_{s2}}{Y_{s1}} = b\log\frac{X_{s2}}{X_{s1}}$$

$$\text{When } \frac{X_{s2}}{X_{s1}} = 10, \qquad b = \log\frac{Y_{s2}}{Y_{s1}}$$

$$\text{If } X_{s1} = 1\gamma \text{ or } 1 \text{ mg.}, \qquad \log X_{s1} = 0$$

$$\log\frac{Y_u}{Y_{s1}} = b\log\frac{X_u}{X_{s1}} = \log\frac{Y_{s2}}{Y_{s1}}\cdot\log X_u$$

$$\log X_u = \frac{\log Y_u - \log Y_{s1}}{\log Y_{s2} - \log Y_{s1}}$$

In a special case, $\frac{X_{s2}}{X_{s1}} = 10$, $X_{s1} = 1$, and $b = 0.333$, when the reaction is measured in mm² as shown by equation 3, the activity of an unknown in reference to a standard is equal to the third power of the ratio of their tuberculin reactions

$$\log\frac{Y_u}{Y_{s1}} = b\log X_u, \qquad X_u = \left(\frac{Y_u}{Y_s}\right)^{\frac{1}{0.333}} = \left(\frac{Y_u}{Y_s}\right)^3$$

Quantitative assay procedure: The activity of a tuberculin preparation of unknown potency may be determined by experiments planned in factorial designs and evaluated through the analysis of variance. The statistical technique adopted here is that of Bliss, which has been used for the assay of penicillin and other drugs (9). The data obtained with a PPD preparation are given in table 2. Four concentrations were used, two designated as "standards" and the other two as "unknowns." The symbols in the table are self-explanatory. The relative potency of the unknown to the standard is obtained by a relative log-potency of $M = \frac{I\,T_1}{T_2}$, where I = logarithm of the ratio of the high dose to low dose for both standard and unknown, a required condition in the assay procedure. In some of the experiments, I was chosen by a ratio of $\log\frac{3.16}{1}$ or 0.5; above equa-

tion thus becomes

$$M = \frac{T_1}{2T_2}$$

The next step is to calculate the standard error of potency. An average standard error is

$$\text{S.E. of potency} = 2.30 \times Sm \times (\text{antilog } M), \text{ where}$$

$$Sm = \sqrt[\lambda]{\frac{1}{N}\left(1 + \frac{T_1^2}{T_2^2}\right)} \quad \text{and} \quad \lambda = \frac{S}{b}$$

(S = standard deviation of the population and b = slope of the regression line)

$$S = \sqrt{\frac{\Sigma(Y^2) - (T_1^2 + T_2^2 + T_3^2)/N}{12(N - 1)}}$$

($\Sigma(Y^2)$ = sum of Y^2S and N = the number of tests in the assay). The slope is calculated by

$$b = \frac{T_2}{2IN}, \qquad b = \frac{T_2}{N} \text{ when } I = 0.5$$

The accuracy of the method is shown by the figures computed from the data of 3 experiments in tables 2 and 3.

The "unknowns" have assayed potencies 148.0 ± 11.95, 181.7 ± 20.0 and 142.7 ± 12.75 per cent that of the standard, while the actual potencies are 150, 200 and 150 per cent, respectively.

As indicated by the dose-response curve in figure 1, along a limited range, the response is approximately linear to the log-dose. Therefore, within this range, the above method may be used by a response-log dose relationship (instead of log-response and log-dose). As may be seen by comparing the values in table 2, there is no gain in the sensitivity of the method by using ordinary numbers instead of logarithms. The use of logarithms for responses has the advantage in that the linear relationship covers the entire range of doses.

Regarding the design of the experiment, the following conditions are suggested for a simple, rapid, and accurate procedure for the assay of tuberculin preparations.

Reactivity: As in any biological assay, the sensitivity and accuracy of a method depend upon both the reactivity and the uniformity in response of the testing animals. In tuberculin assays, therefore, only good reactors should be used. This requirement may usually be fulfilled by using "sound" guinea pigs in which a high degree of tuberculin sensitivity has been established.

Number of determinations: Ten determinations for each concentration, or 40 determinations for each assay, are appropriate numbers required. Forty determinations may be carried out in 5 pigs, by inoculating 4 doses in each side of the dorsal skin.

Dosage: Large doses, but under the necrotizing level at twenty-four hours after inoculation, should be used. The larger the erythema, the less will be the error introduced in reading the skin reaction. One to five γ (0.001–0.005 mg.)

TABLE 2

Assay of Tuberculin (PPD)

(A) $^*U_2$, U_1, S_2, S_1 = 4.74, 1.50, 3.16, 1.00 γ

TEST NUMBER	DIAMETER OF ERYTHEMA (LOG) FOR				$D_1 = U_2 - S_2$	$D_2 = U_1 - S_1$	$D_3 = U_2 - U_1$	$D_4 = S_2 - S_1$	$Y_1 = D_1 + D_2$	$Y_2 = D_3 + D_4$	$Y_3 = D_1 - D_2$
	$^*U_2$	U_1	S_2	S_1							
1	1.08	0.86	1.02	0.85	0.06	0.01	0.22	0.17	0.07	0.39	0.05
2	1.28	1.15	1.20	1.04	0.08	0.11	0.13	0.16	0.19	0.29	−0.03
3	1.22	1.00	1.11	0.98	0.11	0.02	0.22	0.13	0.13	0.35	0.09
4	1.23	1.10	1.18	1.06	0.05	0.04	0.13	0.12	0.09	0.25	0.01
5	1.16	1.06	1.13	1.00	0.03	0.06	0.10	0.13	0.09	0.23	−0.03
6	1.28	1.13	1.20	1.04	0.08	0.09	0.15	0.16	0.17	0.31	−0.01
7	1.31	0.98	1.23	0.90	0.08	0.08	0.33	0.33	0.16	0.66	0.00
8	1.18	1.02	1.13	0.93	0.05	0.09	0.16	0.20	0.14	0.36	−0.04
9	1.20	0.95	1.16	0.85	0.04	0.10	0.25	0.31	0.14	0.56	−0.06
10	1.18	1.02	1.15	0.95	0.03	0.07	0.16	0.20	0.10	0.36	−0.04
Total						1.28 = T_1		3.76 = T_2			−0.06 = T_3

M = 0.1702 or 148.0%; Actual = 150%.
b = 0.376, S = 0.0425, Sm = 0.03774, S.E. of Potency = 11.95%.

(B) $^*U_2$, U_1, S_2, S_1 = 4.74, 1.50, 3.16, 1.00 γ

TEST NUMBER	DIAMETER OF ERYTHEMA (LOG) FOR				$D_1 = U_2 - S_2$	$D_2 = U_1 - S_1$	$D_3 = U_2 - U_1$	$D_4 = S_2 - S_1$	$Y_1 = D_1 + D_2$	$Y_2 = D_3 + D_4$	$Y_3 = D_1 - D_2$
	$^*U_2$	U_1	S_2	S_1							
1	12.0	7.5	10.5	7.0	1.5	0.5	4.5	3.5	2.0	8.0	1.0
2	19.0	14.0	16.0	11.0	3.0	3.0	5.0	5.0	6.0	10.0	0.0
3	16.5	10.0	13.0	9.5	3.5	0.5	6.5	3.5	4.0	10.0	3.0
4	17.0	12.5	15.0	11.5	2.0	1.0	4.5	3.5	3.0	8.0	1.0
5	14.5	11.5	13.5	10.0	1.0	1.5	3.0	3.5	2.5	6.5	−0.5
6	19.0	13.5	16.0	11.0	3.0	2.5	5.5	5.0	5.5	11.5	0.5
7	20.5	9.5	17.0	8.0	3.5	1.5	11.0	9.0	5.0	20.0	2.0
8	15.0	10.5	13.5	8.5	1.5	2.0	4.5	5.0	3.5	9.5	−0.5
9	16.0	9.5	14.5	7.0	1.5	2.5	6.5	7.5	4.0	14.0	−1.0
10	15.0	10.5	14.0	9.0	1.0	1.5	4.5	5.0	2.5	9.5	−0.5
Total						38 = T_1		107 = T_2			5.0 = T_3

M = 0.1542 or 142.7%; Actual = 150%.
b = 10.70, S = 1.237, Sm = 0.03886, S.E. of Potency = 12.75%.

of PPD have been found to be satisfactory doses. The one γ (PPD) dose is chosen so that the units of tuberculin potency may be expressed, if desired, in simple terms of the weights of a standard.

For an "unknown" tuberculin preparation, its potency may be first estimated

approximately with one dose of the unknown and one dose of a standard, and calculated by the third power of the ratio of their skin reactions.

Measurement: The maximal skin reaction, twenty-four hours after inoculation, is favored for the evaluation of tuberculin preparations. It is immaterial whether the reaction is measured by the area or by the diameter of the erythema. For time saving in calculations, the diameter is preferred.

TABLE 3

Assay of Tuberculin (PPD)

(A)

TEST NUMBER	DIAMETER OF ERYTHEMA (LOG) FOR			
	*U_2	U_1	S_2	S_1
1	1.04	0.85	0.95	0.88
2	1.15	0.81	1.02	0.70
3	1.15	1.00	1.10	0.93
4	1.11	0.88	0.98	0.88
5	1.08	0.78	0.98	0.74

Total....T_1 = 0.69, T_2 = 2.09, T_3 = 0.31

M = 0.230, or 170%; Actual 200%
b = 0.299, S = 0.0566, Sm = 0.092
S.E. of Potency = 35.97%

(B)

TEST NUMBER	DIAMETER OF ERYTHEMA (LOG) FOR			
	*U_2	U_1	S_2	S_1
1	1.04	0.85	0.95	0.88
2	1.15	0.81	1.02	0.70
3	1.15	1.00	1.10	0.93
4	1.11	0.88	0.98	0.88
5	1.08	0.78	0.98	0.74
6	1.06	0.78	0.88	0.74
7	1.10	0.85	1.02	0.78
8	1.13	0.88	1.02	0.85
9	1.08	0.88	1.04	0.74
10	1.16	0.90	1.04	0.74

Total....T_1 = 1.66, T_2 = 4.48, T_3 = 0.40

M = 0.259 or 181.7%; Actual 200%
b = 0.320, S = 0.0469, Sm = 0.048
S.E. of Potency = 20.0%

*U_2, U_1, S_2, S_1 = 1, 0.2, 0.5, 0.1 γ of PPD.

SUMMARY

An analysis of the dose-response characteristics of the skin tuberculin reaction has been presented. Expressed in logarithms, the dose and the size of the erythema follow a linear relationship. The response, in terms of the area of the skin reaction, is proportional to the cubic root of the dose of tuberculin.

Based upon the lineality between dose and response, a statistical procedure has been used for the assay of tuberculin preparations. The relative potency of an unknown is computed from the regression line of a standard preparation. The relative potency is determined in experiments planned in factorial design and evaluated through the analysis of variance.

SUMARIO

La Cutirreacción a la Tuberculina para la Valoración de la Tuberculina en los Cobayos

Preséntanse un análisis de las características de la dosis-respuesta de la cutituberculinorreacción. Expresada en logaritmos, la dosis y el tamaño del

eritema mantienen una relación lineal. La respuesta, en términos del área de la cutirreacción, es proporcional a la raíz cúbica de la dosis de tuberculina.

Basándose en la alineación entre dosis y respuesta, se empleó un procedimiento estadístico para la valoración de las preparaciones de tuberculina. La potencia de una incógnita se computa por la línea de regresión de una preparación tipo. La potencia relativa es determinada en experimentos trazados en términos factoriales y valuados por el análisis de la variación.

Acknowledgment

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INHIBITION OF THE TUBERCULIN TYPE REACTION BY ANTIHISTAMINIC DRUGS AND RUTIN[1]

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INTRODUCTION

Since the outstanding elementary experiment of Robert Koch on the reinoculation of tubercle bacilli into the skin of the laboratory animal, much work has been expended on tuberculin sensitivity.

Sensitivity in general has been divided into two groups, humoral and cellular. In the humoral group the circulating antibodies are present in the intercellular fluid and blood stream. In this type of sensitivity the skin responds to intracutaneous injections of offending antigens with an immediate whealing reaction, as, for example, in ragweed hay fever. In the cellular group, the antibodies are present in the cells themselves and the skin responds with a delayed inflammatory reaction. This is the group to which the tuberculin type of sensitivity belongs.

Two well-known methods are used to test for delayed tuberculin sensitivity. The first is the contact or patch test method. When tuberculin is brought in contact with the sensitive skin, a reaction occurs in the epidermis within forty-eight hours with the development of spongiosis and intradermal vesiculation. The second type is the method of the intradermal injection of tuberculin. A reaction develops in the sensitive cutis in forty-eight hours with lymphocytic and epithelioid infiltration and a connective and vascular tissue response.

With the recent development of the antihistaminic drugs, more interest has been displayed in Lewis's histamine theory of sensitivity. It is believed that in the humoral group the union of antigen and antibody causes the release of histamine or a histamine-like substance which gives an immediate reaction. Antihistaminic drugs will inhibit this reaction (1, 2, 3). Histamine, according to present concepts plays no part in the delayed cellular reaction. Inference has been made, therefore, that the antihistaminic drugs have no influence on tuberculin sensitivity. If they do, it would be by some other mechanism.

In order to test further the validity of these theoretical considerations, an attempt has been made to inhibit the tuberculin type of reaction by means of antihistaminic agents and rutin.

METHODS

Old Tuberculin, Tuberculin, Purified Protein Derivative (PPD)[2], the Tuberculin Patch Test (Vollmer)[3], rutin[4], and pyribenzamine hydrochloride[5], in tablet and powder form were used in the following experiments.

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[1] From the Department of Medicine, Section of Allergy, School of Medicine, University of Pittsburgh, Pittsburgh, Pennsylvania.

[2] Parke, Davis & Co.,

[3] Lederle Laboratories.

[4] Generously supplied by E. R. Squibb & Sons, New York, N. Y.

[5] Generously supplied by Ciba Pharmaceutical Products, Inc., Summit, N. J.

Dilutions of Old Tuberculin were made in sterile buffered saline with the same vial of Old Tuberculin. All solutions were freshly prepared before administration, using sterile buffered saline as the diluent unless otherwise stated.

All tests were performed in corresponding sites of the flexor surfaces of the forearm, generally in the middle third. An ordinary 1.0 cc. tuberculin syringe and 25 gauge hypodermic needle were used and all injections were performed and results read by the same observer.

RESULTS

Oral Pyribenzamine and the Tuberculin Test

A group of 29 patients not receiving medication were injected intradermally with 0.1 cc. of Old Tuberculin in a dilution of 1:10,000. Reactions were observed in forty-eight hours, the diameters of swelling and erythema measured and traced on onion skin paper. One week later, these patients began oral administration of pyribenzamine in a dosage of 400 mg. daily in 4 equally divided doses. One hour after the second dose on the second day, when a total of 600 mg. of pyribenzamine had been consumed, the patients were reinjected in the opposite forearm with the same dose of Old Tuberculin. In forty-eight hours, after administration of 1,400 mg. of the drug, reactions were again observed, measured, traced, and compared with the original reactions. The diameter of swelling and erythema was used as the basis for comparison.

Ten patients had identical reactions in both tests, while 4 showed a smaller, and 15 a greater, reaction during administration of pyribenzamine.

Intradermal Pyribenzamine and the Tuberculin Test

One-tenth cc. of 1 per cent pyribenzamine was injected intradermally to produce a small wheal. No subjective symptoms resulted from the injection of pyribenzamine in any case. When the swelling had receded in about ninety to one hundred and twenty minutes, the identical site was injected with 0.1 cc. of 1:1,000 Old Tuberculin. For a control an injection of 0.1 cc. of 1:1,000 Old Tuberculin was administered in the opposite arm. Readings were made in forty-eight hours and compared. Of 5 patients, 4 had a lesser reaction in the site previously injected with pyribenzamine, and one greater, as compared to the controls.

A series of patients previously tested with 1:10,000 Old Tuberculin were given 0.1 cc. of 1 per cent pyribenzamine intradermally as in the previous experiment and were then injected with 0.1 cc. of 1:10,000 Old Tuberculin in the same site in two and one-half to three hours when the swelling subsided. In forty-eight hours the reactions were observed, measured, and compared with previous reaction. Four patients reacted to the same degree, 3 less and 3 more severely following injection of pyribenzamine than when the drug was not present in the skin.

Intradermal Pyribenzamine and the Tuberculin Patch Test

In 11 patients, 0.1 cc. of 1 per cent pyribenzamine was injected in each of two areas on the forearm, so spaced that the tuberculin-impregnated sites of the tuber-

culin patches could be applied directly thereon. Control patch tests were placed on the opposite arm. Strips were removed in forty-eight hours and readings were made at that time. All showed definite positive reactions on the control sites. Eight patients had identical reactions where pyribenzamine had been injected and 3 had a smaller reaction. Five patients were observed for forty-eight additional hours, but there was no change except for fading of all reactions at the time of the final reading.

Iontophoresis of Pyribenzamine and Tuberculin Testing

Pyribenzamine hydrochloride was introduced into an area of skin ($3\frac{1}{2}$ by $7\frac{1}{2}$ cm.) of the forearm by means of a standard iontophoresis apparatus (4, 5) using one third milliampere per sq. cm. for five minutes and a 10 per cent solution of pyribenzamine hydrochloride. One hour later, when the primary erythema had subsided, the tuberculin patch was applied to this site. Control patches were applied to the opposite forearm. Both patches were removed in forty-eight hours. Readings and comparisons were made immediately, twenty-four and forty-eight hours later. Of the 4 patients thus tested, 3 reacted identically in all sites and one had a less severe reaction on the iontophorized site.

In another group of patients pyribenzamine was similarly introduced in an area of the forearm ($3\frac{1}{2}$ by $3\frac{3}{4}$ cm.) with the same apparatus, using one half milliampere per sq. cm. for five minutes. One hour later, with subsidence of erythema, the center of the infiltrated area was injected with 0.1 cc. of tuberculin (PPD) strength 1 or 2. A similar injection of PPD was simultaneously made in the opposite forearm. Reactions were measured in forty-eight and seventy-two hours. Of 5 patients given PPD strength 2, similar reactions in both arms occurred in 2, but 3 had more severe reactions in the arms infiltrated with pyribenzamine. One patient was given PPD strength 1, with identical reactions in both sites.

Simultaneous Injection of Tuberculin and Antihistaminic Agents Mixed in Vitro

PPD tablets, strength 1 or 2, were dissolved in 1 cc. of 1 per cent pyribenzamine. One-tenth cc. of this mixture was then injected intradermally in the forearm and 0.1 cc. of the control strength of PPD in sterile buffered saline was similarly injected in the opposite arm. Reactions were read in forty-eight hours. Six patients in whom strength 1 was used showed marked reduction in size of the reaction in the arm in which pyribenzamine and tuberculin were injected. Two of 3 patients given second strength PPD had a similar inhibition of the reaction.

In 2 patients in whom thephorin was used and in 2 with neohetramine instead of pyribenzamine, this mixture likewise gave smaller reactions than controls.

Histoplasmin and Pyribenzamine

One-tenth cc. concentrated histoplasmin was diluted with 9.9 cc. of 1 per cent pyribenzamine in normal saline and 0.1 cc. of the mixture was injected in the forearm. As a control 0.1 cc. of a mixture of histoplasmin in normal saline in a similar concentration was injected in the opposite forearm. Reactions were

observed in all in forty-eight hours. No differences were noted in either arm in 5 patients. One had slightly smaller reactions with the pyribenzamine and histoplasmin mixture.

Rutin and the Tuberculin Test

A group of 5 patients not receiving medication were injected intradermally with 0.1 cc. of Old Tuberculin in a dilution of 1:10,000. Reactions were observed in forty-eight hours, the diameters of swelling and erythema measured and traced on onion skin paper. One week later these patients began oral administration of rutin in a dosage of 160 mg. daily in 4 equally divided doses. On the first day, the patients received 80 mg. of rutin. One hour after the second dose on the second day, when 160 mg. had been consumed, the patients were again injected in the opposite forearm with the same dose of Old Tuberculin. Reactions were read in forty-eight hours, when 480 mg. of the drug had been administered, and were found to be identical with the original results.

Pyribenzamine and the Tuberculin Test in Guinea Pigs

Twelve guinea pigs weighing approximately 300 gm. were innoculated with tubercle bacilli from an actively growing culture. Six of the guinea pigs were given pyribenzamine orally in a dosage of 20 mg. per Kg. of body weight twice daily. The other 6 pigs were given no medication and were used as controls. On the ninth day all pigs were skin-tested on the abdomen with 1:1,000 Old Tuberculin. There was no reaction. On the twenty-fourth day, one control pig had a positve reaction and on the thirtieth day 3 control pigs were positive and one guinea pig receiving pyribenzamine gave a positive reaction. On the thirty-fifth day, 4 controls and one pyribenzamine guinea pig were positive. On the fortieth day one pig of each group died of tuberculosis. By the forty-third day, 4 controls and 2 pyribenzamine pigs were positive. One pig of each group died on the forty-eighth day, and two days later another pyribenzamine pig died. On the fifty-second day all tests were positive in the 4 controls and the 3 pyribenzamine pigs which still survived. The guinea pigs taking pyribenzamine appeared sleepy and lethargic throughout the experiment. In this small series, there appeared to be a slight delay in the development of tuberculin sensitivity in the guinea pigs receiving pyribenzamine. This may be due to a toxic reaction of the drug causing inhibition of the natural antibody formation.

DISCUSSION

Other workers previously have been unable to affect the tuberculin test using various antihistaminic drugs. Boquet (6) found that the early antihistaminic drugs given subcutaneously did not prevent guinea pigs from responding to tuberculin to which they have previously been sensitized. Breton (7) also was unable to prevent a positive Vollmer patch test by previously injecting 2,339 RP into the test site in a method similar to that used in the present experiments. Guy (8) was unable to inhibit the tuberculin reaction in 5 patients given pyribenzamine orally. However, the delayed eczematous type of reaction can be inhibited with pyribenzamine (9, 5).

In the present study it was not possible to influence the Mantoux test with high oral doses of pyribenzamine. Likewise, there appeared to be little effect on both the patch and intradermal tuberculin test when applied over sites previously infiltrated by injection or by iontophoresis with pyribenzamine.

Although statistically quite a few tests showed some inhibition, in most cases this inhibition was of small degree and probably within the normal variations of the test. Tuberculin reactions are notably inconstant. Of 22 patients tested on three occasions by Guy, only 5 showed rather constant results. Furthermore, Sulzberger states that in some cases repeated injections of tuberculin alter the sensitivity, in some increasing it and in others decreasing it. Rich has also observed variations in reactions to tuberculin tests. When tuberculin is mixed with any antihistaminic drug prior to testing, results are consistently diminished. This may be due to a chemical inactivation of the tuberculin by these drugs *in vitro*.

The histoplasmin skin test is comparable to the tuberculin test in that it also gives a delayed type of reaction. When mixed with pyribenzamine *in vitro* and injected, it failed to antagonize the reaction. Histoplasmin may not be affected by pyribenzamine *in vitro*.

It is believed that there is a relationship between capillary permeability and the delayed inflammatory tuberculin type of reaction. Rutin has been used to decrease capillary permeability. It has been shown (10) that rutin protects against anaphylactic shock. In the present study, however, it was not possible to prevent the tuberculin reaction by the oral administration of rutin.

CONCLUSION

The tuberculin type of sensitivity reaction is not affected by antihistaminic drugs or by rutin.

Pyribenzamine did not prevent the development of tuberculin sensitivity in guinea pigs infected with tubercle bacilli, although some delay in this development was noted.

The oral administration of rutin, another drug which has shown some antianaphylactic activity, failed to inhibit the tuberculin reaction.

CONCLUSIONES

Inhibición de la Reacción de Tipo Tuberculínico por Antihistamínicos y Rutina

La reacción de sensibilidad de tipo tuberculínico no es afectada por los antihistamínicos o por la rutina.

La piribenzamina no impidió la aparición de sensibilidad a la tuberculina en los cobayos infectados con bacilos tuberculosos, aunque se notó cierta tardanza en dicha aparición.

La administración oral de otra droga, la rutina, que ha mostrado alguna actividad antianafiláctica, tampoco inhibió la reacción a la tuberculina.

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EDITORIAL

Ten Beds per Death or Eradication, not Reduction, of Tuberculosis

Institutional isolation of tuberculosis has evolved as an incidental by-product of attempts to improve the condition of individual patients. The pioneers of the sanatorium movement, such as Brehmer and Trudeau, thought chiefly of the value of the sanatorium to the patients. But the value of segregation in diminishing the spread of the disease was suggested at a very early date. Koch himself, when announcing the communicability of this disease, advocated the control of tuberculosis through the suppression of the sources of infectious materials. In his classic book, *The Prevention of Tuberculosis*, Newsholme demonstrated, in 1908, that institutional segregation is the predominant factor in the decline of tuberculosis.

In the days when the tuberculosis death rate in the United States was around 200 per 100,000, there were only a few scattered sanatoriums, occupied to a great extent by what were then considered "favorable" cases for treatment. It then seemed visionary and impractical to ask that every patient with tuberculosis be institutionalized throughout the entire period of active disease, or even that patients should receive sanatorium care during the last year of their lives. But gradually, as facilities increased and tuberculosis decreased, the feasibility and desirability of such goals met with recognition.

In 1916 the Legislature of Massachusetts passed a law requiring hospitalization for tuberculosis equivalent to one bed per two annual deaths or one per 50,000 inhabitants (corresponding to a death rate of 100 per 100,000). In his admirable reports on the Framingham experiment a few years later, Armstrong recommended one bed per annual death. This was accepted by the National Tuberculosis Association and advocated for nearly a generation. By 1929 David Lyman recognized that a ratio of one bed for each annual death was too low. In his presidential address in 1940, Doctor H. D. Chadwick set a goal of two beds per death as the minimum requirement for the control of tuberculosis. Two years later, in *The Modern Attack on Tuberculosis*, Chadwick and Pope recommended at least two and one-half beds per annual death.

It was asserted then that States having as many as three beds per death were more than adequately supplied and would have no waiting lists and even some empty beds. As the tuberculosis death rate declined, however, the ratio of beds to deaths increased so that there is now an average of nearly three beds per death throughout the United States and in some communities more than six beds per death may be available and utilized. But this is still far below the ideal of sanatorium care for every patient affected with active tuberculosis throughout the period of infectivity.

Most tuberculosis workers, like Chadwick, have accepted the Framingham findings that approximately ten cases of active tuberculosis may be found in a community for each annual death. Whether really adequate institutional facilities for the total eradication of pulmonary tuberculosis will require five or twenty

beds per death, however, may vary in different communities, depending on the characteristics of the disease and other facilities available. In neglected groups with a high proportion of acute, rapidly progressive tuberculosis and poor facilities for treatment and consequent short duration of disease, as in the American Negro, Indian, and other special groups, the number of active cases which may be found by survey at any one time may be relatively small compared to the number expiring annually. On the other hand, in groups with sluggish, limited disease and abundant facilities for diagnosis and treatment, like World War I veterans, the duration of the disease may be greatly extended and many arrested, quiescent, and chronic cases may be discovered for each annual death.

There are now nearly 45,000 deaths annually in the United States from tuberculosis. About two-thirds of these, or 30,000 deaths a year, occur among the 120,000 beds in hospitals and sanatoriums. The majority of tuberculosis deaths in institutions, however, occur in general hospitals and other institutions which admit, for the most part, patients in advanced and preterminal conditions. About 14,000 deaths per annum occur in the 19,000 beds in special tuberculosis sanatoriums and hospitals, an intramural death rate of about 15 per cent per annum. Thus more than six beds per death are required to supply sanatorium care for the tuberculous patients of the kind who are at present in tuberculosis sanatoriums. It is notorious, however, that our tuberculosis hospitals and sanatoriums are overcrowded by patients with advanced disease. Ten beds for each annual death from tuberculosis would probably be required to provide institutional segregation for every patient with active tuberculosis in the United States.

The example of the American accomplishment in eradicating bovine tuberculosis within two decades of the initiation of an aggressive campaign against that disease points the direction in which we should go. Other countries, timorous and fearful of expense, have already wasted in condemned meat and low milk production far more than would have been necessary to eradicate the disease among their cattle.

Tuberculin tests and mass fluoroscopic and roentgenographic examinations of entire populations and intensive study of contacts of known cases of tuberculosis will reveal nearly all instances of active, "open" tuberculosis with cavity, the form responsible for most of the spread of the disease.

The increased rest made possible by increased bed capacity of sanatoriums and the newer methods of collapse therapy and chemotherapy have greatly improved the prognosis of these cases. A much greater proportion of patients may therefore be expected in the future to seek sanatorium treatment. Increased general education is needed as to the dangers of tuberculosis and the need for segregation. For those who fail to recognize the value to themselves and to their community of such institutional care and segregation, compulsory measures should be universally instituted just as they are today for other recognized communicable diseases.

It may not be feasible in all places to embark immediately upon an ambitious program for the provision of complete institutional care for all patients with active, communicable tuberculosis. But this can be recognized as our goal and any delay in putting it into execution must be understood to carry with it respon-

sibility for a definite number of deaths from tuberculosis which would be preventable if an all-out campaign for complete hospitalization and tuberculosis eradication were to be undertaken.

Before prematurely considering conversion of tuberculosis facilities and forces for combating other diseases, we should make sure that this condition has been really adequately handled. Patients with closed, arrested, and inactive disease, in whom repeated laboratory examinations fail to reveal discharge of tubercle bacilli, may be cared for in dispensaries, or offices, or in their homes. But any patient who is expectorating tubercle bacilli is a potential hazard to his community and under ideal conditions should certainly be segregated where danger of his spreading the disease may be averted. One bed per death was a grand slogan for 1918, ten beds per death or a bed for every patient with active tuberculosis should be our demand for 1950.

EMIL BOGEN

LETTERS TO THE EDITORS

PRECIPITIN TEST FOR CARBOHYDRATE ANTIBODIES IN HUMAN TUBERCULOSIS[1]

To the Editors of the American Review of Tuberculosis:

We wish to report that a precipitin test for carbohydrate antibodies in human tuberculosis has been obtained. The precipitating agent is the pure water soluble carbohydrate, which has been split by hydrolysis from the antigenic lipo-carbohydrate of the tubercle bacillus.

It has long been known that specific antibodies develop in humans and in experimental animals as a result of infection with the living tubercle bacillus, or through immunization with killed organisms.

From each of the fundamental constituents of the tubercle bacillus (proteins, lipids, carbohydrates), fractions have been isolated which have the property of reacting specifically (some tuberculo-carbohydrates to a high degree) with antibodies which are formed subsequent to infection, or to immunization with the killed organisms. Among these fractions, first the protein and then the lipid components were shown to be actively antigenic in inducing antibody formation when injected into normal animals. However, it has been found that these antigenic components have no power to promote the development of acquired resistance in experimental animals.

Three years ago we showed that the lipo-carbohydrate, extracted from a paraffin oil extract of the tubercle bacillus, is antigenic (Science, 1947, *105*, No. 2715). Preliminary observations in animals have indicated that this *antigenic* lipo-carbohydrate (the only one thus far extracted from the tubercle bacillus) does play a part in the mechanism of acquired resistance.

If antibodies play a part in the process of resistance to infection in human tuberculosis, one should not only be able to detect them in the circulating blood, but also to find some correlation between their presence or absence in the sera of tuberculous patients, and the resistance of these patients to infection, as observed clinically.

We were able to detect these antibodies in sera of tuberculous patients by a direct precipitin test, using as precipitating agent the pure carbohydrate (haptene) which was obtained, by hydrolysis, from our antigenic lipo-carbohydrate. As previously reported, this haptene gives precipitation in dilutions as high as one to ten million, with whole sera of some immunized rabbits (Am. Rev. Tuberc., 1947, *56*, 222).

We did not attempt to make any quantitative determination of the titer of antibodies in the human sera of tuberculous cases, either by diluting the sera or by using the precise quantitative method developed by Dr. M. Heidelberger. Thus, our results have only a qualitative value.

Technique of the precipitin test: To perform the test, venous blood from human beings was collected in vacuum tubes, allowed to clot at room temperature, then stored at 4°C. overnight. Serum was removed after centrifugation at 2,000 r.p.m. for fifteen minutes, then recentrifuged at 4,500 r.p.m. for five minutes. Two cc. of sera were put in each of two sterile Wassermann tubes. To one of these tubes antigen was added, the other tube was kept as a control. The amount of antigen necessary to bring about a satisfactory

[1] This study was carried out under a grant from the Josiah Macy Foundation.

precipitation in a positive serum was found to be 5γ of the soluble carbohydrate, *i.e.*, one-twentieth of a cc. of 0.1 mg. per cc. solution. The tested sera as well as the control sera were stored at 4°C. for eight to ten days, and the readings were made after centrifugation for five minutes at 4,500 r.p.m. In a positive serum, a firm transparent disc is usually formed, which is easily seen by slight agitation of the tube. A false precipitate is sometimes found in both tubes, but these rapidly go into solution after some agitation.

Results: Two hundred ninety-eight sera from cases of tuberculosis have been tested thus far through the collaboration of Dr. A. Chaves. The classification of the state of the disease for all cases was made by Doctor Chaves. The specimens were obtained from various Health Department clinics and several hospitals. Two hundred nine of the 298 specimens were obtained from cases of active tuberculosis, comprising all varieties of the disease from rapidly progressive cases to regressive cases. Eighty-nine were inactive cases, classified according to whether the disease had been present for less or for more than two years. A table of the results obtained in the various groups is presented. The total in each group is small, but the trend of the results is interesting.

	TUBERCULOUS CASES					NORMAL AND OTHERS
	Active			Inactive		
	Rapidly progressive	Intermediate	Regressive	0 to 2 years	Over 2 years	
+	23	36	37	22	5	5
−	50	39	24	17	45	94
Number	73	75	61	39	50	99

It will be noted that antibodies were not found in the blood of all the active cases. Only in about 50 per cent of the active cases were the sera found to be positive. Nevertheless, the relative proportion of the positive to the negative sera was three times higher in the regressive cases (column 3) than in the rapidly progressive cases (column 1). An almost equal number of positive and negative sera was found in the intermediate group.

Antibodies were found in some of the inactive cases, but the relative proportion of positive to negative sera is very much higher in the infections which had been inactive for less than two years than in those inactive for more than two years.

As a control for the specificity of this precipitin test, the sera of 99 persons who presumably did not have tuberculosis were examined. Included in this group were specimens obtained from 47 normal persons, 5 patients with syphilis plus a pool of sera from 10 other early syphilitic infections, 13 patients with pneumonia, 15 with cancer, and 18 who had various other illnesses. Five positive reactions were found in this group of 99 individuals. One person was a contact of a case of active tuberculosis, 2 had recently recovered from a nontuberculous pneumonia, and 2 were patients in the chest clinic who were classified as "normal." The diagnosis of "nontuberculous" was made only on the basis of the chest roentgenogram.

These results seem to indicate that carbohydrate antibodies are more prevalent in tuberculous patients who are successfully fighting against the disease than in patients in which the infection is progressing rapidly. The results also show that antibodies remain present for a certain period of time in the sera of patients who have overcome the disease (from the clinical point of view).

This preliminary exploration suggests a possible correlation between these carbohydrate antibodies and the acquired resistance to the tuberculous infection. Quantitative serial determinations of the titer of antibodies in selected cases are needed before a conclusion can be reached concerning the protective power of these carbohydrate antibodies in human tuberculosis. The fact remains that a specific precipitin test is, for the first time, available in this disease.

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March 26, 1919.

BOOKS

Tuberculosis in the British Zone of Germany, with a Section on Berlin. Report of an Inquiry Made in September–October 1947 by M. Daniels, M.D., D.P.H., and P. D'Arcy Hart, M.D., F.R.C.P., Members of the Scientific Staff Medical Research Council. Pp. 32, His Majesty's Stationery Office, London, 1948, paper, Sixpence net.

Report on Tuberculosis in Germany (U. S. Zone) by a Commission Appointed by the Secretary of the Army, Composed of Esmond R. Long, M.D., Philip E. Sartwell, M.D., Colonel Silas B. Hays, M.D., and Major Alonzo W. Clark, M.S.C., March 5, 1948. Pp. 37.

Under government auspices both Great Britain and the United States have made surveys in occupied Germany for the purpose of determining the influence of World War II on the incidence, course, and death rate of tuberculosis during the War and its subsequent course and prevalence. The writers of these reports also discuss discovery, reporting, prevention, and management at the present time and present a discussion of manifest trends.

The assignment of the British investigation arose through a request from the Public Health Adviser addressed to the British Military Governor. As a result, the Ministry of Health was invited by the Foreign Office to arrange the investigation. The investigations were carried on in September and October, 1947, and reported in 1948. The main assignment was to the British Zone of Germany but attention was also directed to the situation in Berlin.

The survey conducted by the United States was initiated by orders from the Secretary of the Army, dated February 9, 1948. The members of the United States commission were directed "to investigate the incidence of, and recommend control measures for, tuberculosis among the German civilian population." The investigations were carried out and reported under date of March 5, 1948. This survey was limited to "Berlin and the three largest Länder of the U. S. Zone."

The British and the U. S. teams devoted approximately the same length of time to the respective surveys. Though not identically stated, their objectives were virtually the same, the problems confronting them were almost identical, their methods of approach were similar, and their findings were so nearly parallel as to inspire added confidence in their findings. In this brief review of the two reports, we can mention only their principal findings in relation to the effects of war and postwar trends.

Both surveys show the upsurge in the mortality rates as the War progressed, reaching its peak in 1945 and gradually declining since the War. On the whole, the rise in mortality was decidedly greater in urban than in rural communities and the larger the city, the sharper the rise. It is thought particularly by the U. S. commission that bombing, destruction of homes, the interruption of communications, and the resulting nervous strain plus a shortage of food supplies were important contributing factors. The British report indicates that in Berlin the death rate from all forms of tuberculosis was 90 in 1937 and 291 in 1945 (the peak). The United States study arrived at virtually identical conclusions. Both reports indicate that local health authorities through case finding and reporting are of the opinion that the incidence of active tuberculosis is increasing but the reporters believe this is a consequence of revival of the clinical services and case finding facilities and improved methods of diagnosis and discovery.

There is uniformity of opinion concerning the relatively high bed capacity for the management of tuberculous cases except in the city of Berlin. The situation in the British

Zone is decidedly better than in Great Britain. Compared on the basis of the usual ratio of beds to annual tuberculosis deaths, the report shows 2.2 in the British Zone as compared to 1.3 in England and Wales. In the U. S. Zone, approximately 2,000 beds are needed to bring the ratio up to two beds per annual tuberculosis death and 8,800 beds would be required in Berlin to meet this standard. The British report discusses mass roentgenographic findings which seem not to be particularly out of line with such surveys in England and America. The United States report mentions the employment of BCG and approved its employment provided it did not lessen the emphasis on other methods of control. It is agreed that in both the British and the U. S. Zones there seems to be a moderate postwar increase in the nonpulmonary forms of tuberculosis and that this may be attributed to the high incidence of tuberculosis among the dairy herds and imperfect methods of pasteurization of the milk.

Both reports are well supplied with tables and charts. The U. S. report covers World War I, thus posing interesting questions and permitting valuable comparisons. According to the United States report, the immediate effects and the ultimate course seem to run virtually parallel in the two World Wars and the figures confront us with a sharp warning against World War III.

In the opinion of the reviewer, these reports clearly show the evil effects of the stress and strain of war, both psychic and somatic, including disturbed nutrition, destruction of normal habitations, and the resulting displacements. These reports merit a careful study by all who are interested in tuberculosis.

LEWIS J. MOORMAN

WILLIAM B. TOLLEN: *Irregular Discharge: The Problem of Hospitalization of the Tuberculous, with a Foreword by Paul B. Magnuson and a Preface by John B. Barnwell. Veterans Administration, Washington, D. C., 1948. 64 pp. pamphlet, available from the Superintendent of Documents, Washington, D. C.*

The Veterans Administration operates, among other things, the largest single hospital system in the world for the treatment of tuberculous patients. It should provide a peculiarly favorable laboratory for the study of some of the unsolved problems in the control of this disease and the treatment of such patients. To some degree its tuberculous patients are a selected group in that they are predominantly males and that at some time they have been selected for military service. The net result, however, is a patient group far more homogeneous than any group of similar number which has been studied or may be studied in terms of behavior and its bearing on recovery. Mr. Tollen has utilized this unique laboratory to study irregular discharge which is a generic term for the inability or unwillingness of patients to complete medically indicated hospitalization.

As the author points out, veterans' hospitals enjoy no monopoly of this problem. The literature on this subject from which he cites, while scanty in quantity, is sufficient to indicate that the rejection and premature termination of prescribed treatment seriously limits many preventive and therapeutic efforts. It would appear that isolation can aid tuberculosis control only when it consistently and successfully isolates a substantial proportion of infective patients. In many resistant groups, younger adult males are often conspicuous among the problem cases.

In a number of previous studies the data, as above suggested, are sometimes incompletely homogeneous. Some of them are colored by the a priori convictions of the authors who, in some instances, have been a party to such conflicts over long periods of time.

Often these writers' knowledge of the disease has seemed far superior to their grasp of the intricacies of human behavior.

Possibly as a result of reviewing such studies in advance of this study, a fresh approach and technique have been utilized. Experienced social workers, to whom the assignment was a new task and not an ancient frustration, were assigned to interview veterans whose discharge had been irregular. The sample selected was the accessible portion of all irregularly discharged patients who left treatment facilities during July, 1947. The hospitals were operated or financed by the Veterans Administration. The number of patients interviewed was 401.

This procedure yielded some figures which have been presented in appropriately simple tables. In this study they are used to check and validate qualitative observation and experience rather than as an isolated source for conclusions. The selection of a single time interval eliminates variations due to changing economic and seasonal factors. The author has carried on concurrently a thoughtful examination of medical and psychological literature dealing with the behavior of patients and others in comparable circumstances. Accordingly, this reviewer is perplexed that Mr. Tollen appears to remain unaware of the degree to which chronic patients other than the tuberculous also resist medical intentions and advice. The problem of infection has brought such behavior into the spotlight in tuberculosis. Are we to believe that the death toll from coronary thrombosis, for example, includes no persons who have turned their backs on sound medical advice? Or are we dealing with a common human pattern of behavior which the presence of tuberculosis throws into high relief?

In this report we have presented to us the statements of the patients regarding their motivation and evaluation by trained interviewers, who also had access to hospital records. Whether or not one agrees wholly with such evaluation, it is apparent that the study is an effort to ascertain the facts and not an attempt to justify (or to discredit) medical or administrative method. The willingness of the Veterans Administration to review its effectiveness in this way is a reassuring attitude. The efforts of medical personnel to supplement its own perspectives by those of other disciplines also augurs well for the future development of the VA Tuberculosis Service.

The author's conclusions, which he presents with considerable eloquence, are far from arbitrary. He offers no single panacea. On two points his conclusions resemble those of Betty MacDonald (author of *The Plague and I*, a lay study of how tuberculosis treatment looked to a patient), first that recovery from tuberculosis may depend considerably upon the degree of self-discipline achieved by the patient and, second, that the confidence of the patient in the persons who order and administer his treatment may be a key to that discipline. Both authors reject the notion that everything beside the lesion may be dumped into the lap of some extra-medical worker and dismissed, no matter what skills are involved. Both see the most complete supplemental staffing of a hospital as but the lengthened shadow of the physician and his understanding of the patient's problems of treatment and recovery.

It is to be hoped that this pamphlet is only the first installment in a habitual and continuing study of the behavior of tuberculous veterans and its relation to their treatment and recovery. This publication is unimpeded by professional jargon or official gobbledegook. It provides food for thought both for those who are concerned with veterans' affairs and for those who wish to make isolation and treatment of the tuberculous a more effective instrument in the control of the disease.

HOLLAND HUDSON

H. BURNAND, H. JAEGER, M. AMSLER, F. VERREY, E. MARTIN, J. L. NICOD, P. HARDUROY: *Le Problem des Tuberculoses Atypiques. Pp. 429 with 89 illustrations, Masson et Cie., Paris; F. Roth et Cie., Lausanne, 1946, Paper.*

This volume is not merely a discussion of unusual forms of disease known to be caused by the tubercle bacillus. It also treats of conditions which have only been suspected of such an origin. The problem of determining from analogy and deduction, with a reasonable degree of probability, the tuberculous nature of an affection, without demonstrating the actual presence of the Koch bacillus in its lesions, is a difficult one. In this work, seven Swiss scientists from the universities of Lausanne and Zurich and from Geneva have attacked it valiantly and comprehensively. Their avowed purpose has been to assemble in a manual the current ideas about undeveloped and latent tuberculosis with such clinical, histological, and bacteriological evidence as is available to support them. That they have been able to produce and publish so readable and useful a volume so soon after the war is to their credit. This reviewer does not know of any similar work in English.

In their clinical studies they have investigated atypical tuberculoid conditions in the lung, including labile pulmonary infiltrates such as are seen in epituberculosis, in Löffler's syndrome, in certain cases of asthma in bronchiectasis, in a condition they designate as "debility of the bronchial mucosa" and in Boeck's sarcoid. An entire chapter is devoted to conditions attributed to the action of toxins derived from a tuberculous focus, possibly remote or concealed. Such conditions are often diagnosed as due to constitutional inadequacy. The authors consider them to be the result of a state of chronic infection or of latent tuberculosis. Such conditions are perhaps more readily attributed to tuberculosis in Europe than they would be in localities in America where tuberculosis is less prevalent.

Dermatological and ocular conditions suspected of having a tuberculous origin are fully discussed as is "tuberculous rheumatism" of which so much is written in French and so little in English.

Nicod in his summary of the histological studies speaks of early nonspecific changes due to tuberculosis which never reach the stage of tubercle formation but remain simple inflammatory reactions. On the other hand, typical tuberculous processes, such as cascation or calcification, may eventually disappear in healed or latent tuberculosis and be replaced by fibroid or cicatricial lesions. Harduroy, the bacteriologist, discusses atypical forms of the tubercle bacillus. Burnand sums up the results of all these studies in a paragraph of "final considerations" instead of "conclusions." Identification of acid-fast tubercle bacilli, he says, can no longer be considered the sole criterion of atypical tuberculosis. While we wait for new laboratory techniques, lacking today, there are certain morbid states that can be recognized as tuberculous by their clinical manifestations. This is in line with the present trend of considering the inherited and acquired characteristics of the individual as prime factors in the development and course of his disease.

ARTHUR T. LAIRD

SELMAN A. WAKSMAN. *The Literature on Streptomycin 1944–1948. Pp. xv–112, 1948, Rutgers University Press, New Brunswick, New Jersey, cloth, $3.00.*

A bibliography of the world's literature on streptomycin is of great value to both clinicians and research workers. This small volume lists 1171 papers, monographs, and books dealing with every aspect of the subject. Another 40 references are given to work on the

Actinomyces, the production of antibiotic substances, and on streptothricin. The author index has cross references to all the authors of each paper and is very useful. The references on streptomycin, however, are listed roughly in the order of their appearance and are indexed only by the principal topic of each paper. This arrangement detracts considerably from the usefulness of the volume, especially for those who are not already familiar with much of the work on streptomycin. The important paper which appeared in 1944 announcing the isolation of streptomycin by A. Schatz, E. Bugie, and S. A. Waksman is reprinted in full.

LAWRENCE B. HOBSON

J. K. DONALDSON: *Surgical Disorders of the Chest. Second Edition. Pp. 485, Lea and Febiger, Philadelphia, 1947, cloth, $8.50.*

The main additions to the first edition of this work (1944) include a description of the advances made in traumatic thoracic surgery during World War II and more details in the treatment of pleurisy, surgical diseases of the esophagus, and the surgery of congenital cardiovascular anomalies.

In his preface, the author states that the volume is primarily designed as a reference book for physicians, surgeons, and medical students. He indicates that sufficient technical detail is included to permit the competent general surgeon to treat certain surgical disorders of the chest.

As the most recently published attempt to correlate what is new in the surgery of the chest, the book is certainly worthy of serious consideration and careful study. Many medical students and physicians, not specialists, will find it a useful reference text. The inclusion of the chapter on *Resuscitation and Inhalation Therapy* is a welcome addition to a surgical volume.

On the critical side several features of the book must be discussed. The over-all impression is that the text was written hurriedly. There are far too many references to other sections in the book, particularly following sections. This, combined with frequent parenthesized directions, makes for poor continuity both of text and thought. As a consequence either of carelessness in writing the original text or inadequate proof-reading, there are at least 40 examples of misspelled words, unwieldy English, and misquoted statistics.

Frontal roentgenograms are not positioned uniformly. On pages 132 and 310 they are reproduced differently in the same figure. On page 222 the roentgenogram is upside down. The roentgenographic illustrations frequently are inadequate, particularly the bronchograms. This may be due in part, however, to a poorer grade of paper in the second edition, undoubtedly necessary because of war restrictions.

In general, the illustrations are adequate and many are excellent. Figure 107 (page 324) is however, extremely small and is confusing even to a surgeon with considerable knowledge of hilar anatomy. Unfortunately, the colored plates on decortication lack third dimensional detail.

A few of the more obvious deficiencies in the text should be listed. The important role of endoscopic procedures in diagnosis and treatment is nowhere emphasized sufficiently. Specific examples are found in the inadequate discussion of indications for bronchoscopy in bronchiectasis, postoperative pulmonary complications, and tuberculosis. The inference is made throughout that the thoracic surgeon usually does not perform his own endoscopies, an entirely unwarranted impression.

On page 156, in discussing the treatment of tuberculous effusion and empyema (which this reviewer believes would preferably be discussed with tuberculosis) no mention is made of the status of the underlying lung as a factor in dictating either treatment or prognosis. On page 256, there is an inadequate discussion of pulmonary hilar and segmental anatomy. On page 291, only passing comment is made on carcinoma of the upper esophagus and there are no references in the bibliography to carcinoma in this location. The description of pulmonary cysts, emphysematous blebs and their differential diagnosis on page 306 is inadequate. There is insufficient separation of so-called bronchial adenoma from primary carcinoma of the lung (page 312). Specifically, there is no description of the microscopic pathology and no attempt at clinical differentiation. On page 361 *et seq.*, the collapse therapy of pulmonary tuberculosis is covered rather sketchily. I disagree with the expressed opinion that the objective of penumothorax therapy is complete collapse of the lung. Streptomycin therapy is not discussed at any length, but the section may have been completed too early for attempts at critical evaluation. There is no discussion of open pneumonolysis or of pneumoperitoneum. The section on extrapleural pneumothorax belongs in the chapter on extrapleural pneumonolysis. There should be greater elaboration of the role of lobectomy and pneumonectomy in the treatment of pulmonary tuberculosis.

Finally, a discussion and description of methods which are now obsolete add nothing to the practicability of a book designed primarily as a working manual for others than thoracic specialists. Only a few of these will be mentioned. "Emergency" pericostal suture for a bleeding intercostal vessel is a discarded technique. The use of an indwelling needle in the treatment of pressure (not tension) pneumothorax is not the best practice. There is no longer a necessity for stressing the avoidance of entering the pleural cavity in the surgical approach to cardiac wounds, or in pericardectomy. There is little or no object in describing an extrapleural approach to posterior mediastinal tumors. The text contains confusing allusions to tourniquet ligation in lobectomy and pneumonectomy. The inference that tourniquet ligation is *ever* indicated in pneumonectomy for carcinoma is challenged.

In conclusion, the rapid improvement of thoracic surgical techniques and the widened indications for operative interference in many thoracic diseases make it advisable that this material be assembled in available form. The author obviously has expended much effort in the preparation of the first and second editions. It is to be hoped that, if a third edition is contemplated, more care will be exercised in obtaining smoothness and clarity; that pertinent additions and deletions will result in a more useful compendium.

PAUL C. SAMSON

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